SOME ASPECTS OF GASTRO-OESOPHAGEAL REFLUX IN ANAESTHETIZED SHEEP

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August, 1984

To my mother V. Nesa

and wife Dilruba

I declare that the contents of this thesis are my own work and have not been presented to any University other than the University of Edinburgh.

August, 1984

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ACKNOWLEDGEMENTS

I wish to thank Professor J.R. Campbell for his help and for the facilities made available to me in his department. I am indebted to Mr. M.A. Camburn and Dr. D.F. Cottrell for their invaluable advice and constructive criticism throughout this project.

My thanks to Mr. A. Riley and Mr. W. Wilson for handling the experimental sheep in this study.

It is a pleasure to record my thanks to Mr. R. Munro and Miss F. Manson for the photography. Thanks are also due to Drs. C.M. MacGregor and T. Fernandez for their help and encouragement.

Finally, I would like to thank Mrs. C. Dickson for preparing the typescript.

SUMMARY

Gastro-oesophageal reflux is a serious problem associated with ruminant anaesthesia. The results of the present investigation of this problem are summarised below.

Management of animals prior to and during anaesthesia influenced the incidence of reflux. Depriving the sheep of food and water for 24 hours prior to induction of anaesthesia significantly reduced the incidence of reflux. The occurrence of reflux was also influenced by the positioning of the animal during anaesthesia. Dorsal recumbancy with the head tilted down was the most vulnerable position. In this position reflux occurred in 100% of cases as compared with right lateral recumbancy with head up position where the occurrence of reflux was only 12%.

Intraruminal pressure built up during anaesthesia was monitored over a period of 60 minutes. Two planes of anaesthesia, light and deep, were maintained for two 30 minute periods in a single anaesthetic session. Intraruminal pressure build up in deep followed by light anaesthesia was significantly greater than that in light followed by deep anaesthesia. The maximum pressure gradient between the rumen and thoracic oesophagus and the occurrence of reflux were recorded and it was found that only 24% of refluxes were associated with the maximum pressure gradient. The intraruminal pressure build up in the starved sheep was significantly lower when compared with that of the unstarved sheep.

The effects of intraruminal insufflation on the occurrence of reflux were investigated and it was found that the intraruminal pressure required to produce reflux was unusually high (about 40 mmHg) in comparison with pressures that can be built up during normal periods

of anaesthesia lasting for 2-3 hours.

The influence of halothane anaesthesia on the motor functions of the oesophagus and rumen was studied using manometric and electromyographic techniques in both light and deep planes of anaesthesia. The motility of these organs was frequently present in light anaesthesia while it was almost totally abolished in deep anaesthesia. The oesophageal pressure waves in light anaesthesia were always peristaltic in nature, the velocity being 25 cm/sec.

A zone of high pressure was detected at the gastro-oesophageal junction in the anaesthetized sheep using balloon tip catheter and a pull through technique. The length of this zone was 2.9 cm. The resting LOS pressure measured by balloon tip catheter was significantly greater than that measured by open tip catheter. The resting LOS pressure was not significantly influenced by the depth of anaesthesia. The LOS usually contracted prior to rumen contraction in light anaesthesia. In deep anaesthesia, when the oesophageal and ruminal contractions were totally abolished, the LOS still showed rhythmic fluctuation in baseline pressure.

The response of LOS to an increase in intraruminal pressure was studied. The LOS pressure was also increased with the increase of intraruminal pressure. The occurrence of reflux was associated with diminished LOS pressure.

The influence of pentagastrin, atropine sulphate and propranolol hydrochloride on LOS pressure was studied. The LOS pressures were increased within one minute of pentagastrin injection while after atropine, the pressure was decreased within one minute. These changes in LOS pressure however were not statistically significant.

Propranolol (after atropine) also had no significant influence on LOS pressure.

The motor activities of the oesophagus, gastro-oesophageal junction and reticulorumen were studied by electromyography (EMG). These activities in the cervical oesophagus were characterised by individual spikes in light anaesthesia which were almost abolished in deep anaesthesia. The reticular and ruminal EMGs comprised of regular spike bursts, the reticular bursts usually preceded the ruminal ones. These activities were usually present in light anaesthesia while in deep anaesthesia they were totally abolished. The electromyographic activity of the gastro-oesophageal junction was characterised by continuous spike discharges which were present in both light and deep planes of anaesthesia.

The influence of intravenous anaesthetic agents (pentobarbitone, thiopentone, chloral hydrate-magnesium sulphate and alphaxalone/alphadolone) on the intraruminal pressure build up, oesophageal and ruminal motility and gastro-oesophageal reflux was studied. The largest intraruminal pressure build up was found in alphaxalone/alphadolone anaesthesia and the smallest with thiopentone anaesthesia. The highest incidence of reflux was found with thiopentone (70%) and the lowest with alphaxalone/alphadolone (40%) anaesthesia.

The barbiturates (pentobarbitone and thiopentone) caused total abolition of oesophageal and ruminal motor functions in both light and deep planes of anaesthesia. These activities, however, were frequently present in light chloral hydrate and alphaxalone/alphadolone anaesthesia. The direction of the oesophageal pressure waves in these studies was peristaltic.

CHAPTER ONE

GENERAL INTRODUCTION

Although it is commonly agreed that the major hazard associated with general anaesthesia in ruminants is the problem of regurgitation or reflux of rumen contents, a review of the literature reveals that little is really known about the mechanisms involved. This literary search also indicated that there has been no experimental study to investigate the phenomenon or the mechanism involved.

Inhalation of rumen contents is possibly the most serious accident that can happen during anaesthesia in ruminants (Messervey and Jones, 1956; Hecker, 1974; Boyd and Ducker, 1975; Jones and Prentice, 1975; Laitinen, Mokka, Valanne and Larmi, 1978; Raptopoulos, 1983). Death may rapidly follow from bronchospasm which frequently proves refractory to treatment (Hall, 1971). Where bronchospasm does not cause death, the inhaled ingesta can provoke an aspiration pneumonia (Leek, 1975) that is usually unresponsive to all forms of treatment, again resulting in the death or the humane destruction of the animal. To eliminate this hazard some surgeons have been reported to perform tracheotomies using local anaesthetics before attempting to administer general anaesthetics (Hecker, 1974).

Leek (1975) when considering the possible mechanism of reflux in his paper to the Association of Veterinary Anaesthetists at Cardiff suggested that "There seem to be four circumstances by which rumen contents may gain access to the oesophagus: (i) regurgitation of the cud bolus during rumination, (ii) "vomiting" under certain experimental conditions, (iii) eructation of ruminal gases and (iv) passive reflux of contents through relaxed oeosphageal sphincters". He also suggested that "The anaesthetic agent depresses medullary centres including the

oesophageal centres. In consequence, oesophageal tone is reduced and the sphincters relax. Rumen contents trickle passively into a partially relaxed oesophagus. This passive reflux may be aided by gravitational forces depending on body position, by the development of slight ruminal tympany, by coughing (during, for instance, the introduction of an endotracheal tube) and by bodily movements (e.g. during struggling, repositioning of the body). Under light anaesthesia, swallowing movements do occur and these may or may not clear the oesophagus. Under deep anaesthesia, swallowing movements are absent. There seems to be little correlation between the depth of anaesthesia and the reflux of rumen contents".

Although there has been very little experimental work to study the influence of anaesthesia on the oesophagus and reticulorumen, these organs have been extensively studied in conscious and decerebrate preparations in sheep in developing a knowledge of the functions of the ruminant digestive system (Dougherty and Meredith, 1955; Stevens and Sellers, 1960; Titchen, 1979).

The present study was initiated in an attempt to identify the mechanism or mechanisms involved in the problem of regurgitation or reflux of rumen contents during anaesthesia. Sheep were chosen as the experimental model and as a representative of the ruminant species.

CHAPTER TWO

REVIEW OF THE LITERATURE

INTRODUCTION

Gastro-oesophageal reflux (GOR) is a widely recognized hazard in veterinary and human anaesthesiology. The mechanism underlying GOR has already been investigated experimentally and the aim of the work reported here was to study in detail some aspects of GOR in anaesthetized sheep. In order to understand the probable mechanisms which operate or are modified during anaesthesia it would be helpful to discuss the various physiological processes involved in gastro-oesophageal function which operate in the conscious ruminant animal. A review of the literature must therefore include a description of the anatomy of the region and of the processes of regurgitation, eructation, swallowing and vomiting behaviour for which considerable experimental evidence exists.

Because of the recurring need to describe the region where the oesophagus and reticulorumen (stomach) join, some definition of terms is required. A number of descriptive terms are used throughout the literature. These include lower oesophageal sphincter (LOS), cardia, cardiac orifice, gastro-oesophageal junction, junctional zone and are often used loosely and interchangeably. LOS is a functional description because it represents an area of increased intraluminal pressure. Cardia and gastro-oesophageal junction are often used functionally and are anatomical terms. The term LOS has been used for the description of my own study and the terms used by others are reported as used.

ANATOMY OF THE OESOPHAGUS

Sheep

The oesophagus is a 45 cm long musculomembranous tube. Its diameter at the level of the pharynx is 1.8 cm which increases caudally up to 2.5 cm at the level of cardia (Sisson and Grossman, 1975). The muscular coat has two distinct layers. The direction of muscle fibres is common in all mammals: the outer layer is longitudinal and the inner layer is circular. Muscles of both layers are striated throughout the length of the oesophagus. At the cardia, the skeletal muscles change abruptly into smooth muscles. The overlapping of pharyngeal and oesophageal musculature forms the pharyngo-oesophageal sphincter. The epithelial lining of the oesophagus consists of stratified squamous cells (Sisson and Grossman, 1975).

COURSE AND RELATIONSHIPS

Cervical Part

Course: The oesophagus begins dorsal to the cricoid cartilage of the larynx and passes along the dorsal surface of the trachea to the second cervical vertebra (May, 1970). It then inclines to the dorso-lateral surface of the left side along which it continues to the thoracic inlet where it again moves dorsally.

Relationships: Dorsally - the longus capitis and colli muscles; laterally - the left common carotid artery, left internal jugular vein and the omohyoid and scalenus muscles; ventrally - the dorsal cricoarytenoid muscle, trachea, left common carotid artery and left vago-sympathetic trunk (near the thoracic inlet), and medially - the trachea and right longus colli muscle. The external jugular vein may be

related to the lateral surface near the thoracic inlet (May, 1970).

Thoracic Part

Course: The oesophagus enters the chest and lies to the left of the trachea as far as the second intercostal space (May, 1970). It then inclines dorsally and to the right until it becomes dorsal to the trachea, crossing the bifurcation of the latter in the median plane. From here it passes through the post-cardiac mediastinum to the hiatus oesophagus of the diaphragm, which is about the level of the ninth or tenth thoracic vertebra.

Relationships: To the right - trachea, to the left - the roots of the bracheal plexus, the left costo-cervico-vertebral trunk and the corresponding vein, the anterior duct, the aortic arch and the sympathetic ganglia (May, 1970). It is also intimately associated with the vagus nerve. In the post-cardiac mediastinum, the caudal mediastinal lymph glands lie along its dorsal surface.

Abdominal Part

An intra-abdominal part of oesophagus measuring from 1.0-3.0 cm in length is reported by Winship, Zboralske, Webber and Soergel (1964).

Nervous Innervation

Extrinsic: The oesophagus receives its nerve supply mainly from branches of the vagus nerve (Dougherty, Habel and Bond, 1958).

Nerves	Area of Innervation	Type of Fibre
Pharyngo-oesophageal nerve	Pharyngo-oesophageal sphincter, cranial half of the cervical oesophagus, and in some cases, the entire cervical oesophagus as far caudally as the thoracic inlet.	Motor

Nerves	Area of Innervation	Type of Fibre
Recurrent laryngeal nerve	Cranial part of the thoracic oesophagus	Motor
Cranial laryngeal nerve	The whole of oesophagus	Sensory
Dorsal oesophageal branch of vagus and dorsal vagal trunk	Covering the area from the level of heart to the cardia	Motor

Intrinsic: Reports of intrinsic innervation to the oesophagus of sheep are not readily available in the literature. However, the existing information in other species would be useful for a better understanding of its physiology. The intrinsic innervations to the oesophagus comprise the intramural neurons and their extensions (Goyal and Cobb, 1981). These neurons and extensions form the myenteric plexus which are distributed in the narrow connective tissue layer that separates the muscle layers and are best developed in the lowermost portion of the oesophagus (Botha, 1962; Thomas, 1981). In the oesophagus and lower oesophageal sphincter (LOS) the myenteric plexus is distributed throughout both striated and smooth muscle portions (Ingelfinger, 1958; Mann and Shorter, 1964; Jacobwitz and Nemir, 1969; Thomas, 1981). The neurons of this plexus connect with both postganglionic sympathetic and preganglionic vagal fibre synapses. The final common pathway fibres of the autonomic nervous system are anatomically indistinguishable (Thomas, 1981). The main function of this plexus is to control the motor activities of the oesophagus. But the functions of these intramural plexuses within the striated muscle oesophagus remain obscure since histological studies suggest that the efferent vagal fibres do not synapse on cells of the myenteric plexus, but end directly on the striated muscle cells at neutro-muscular

junctions similar to those of skeletal muscle fibres elsewhere (Samarasinghe, 1972; Floyd, 1973).

Man

The muscular coat consists of striated and smooth muscles. The striated muscle is generally limited to the upper two-thirds of the oesophagus; the lower third contains smooth muscle only (Gray, 1964). In the upper quarter, both layers consist of striated muscle. In the second quarter, bundles of smooth muscles appear and these gradually replace the striated muscle more caudally.

Dog

The entire oesophagus is striated muscle and there is no gross demarcation between the striated musculature of the oesophagus and smooth muscles of the stomach (Miller, 1979).

ANATOMY OF THE RUMEN

The rumen is a bulky muscular sac occupying most of the left half of the abdominal cavity and extends considerably to the right of the median plane (Sisson and Grossman, 1975). The muscular coat of rumen consists mainly of smooth muscle; however, some striated muscle fibres from the longitudinal muscle layer of the oesophagus also radiate over some parts of the rumen (Nickel, Schummer and Seiferle, 1973). The muscular coat of rumen consists of three layers: longitudinal, circular and oblique. The arrangement of these layers however are not uniform.

Innervation

The ruminal innervation in sheep has been extensively studied by Habel (1956). He found that the rumen is supplied almost entirely by the dorsal vagus. The right and left vagi pass over the heart and divide into dorsal and ventral branches, which unite with their counterparts to form the dorsal and ventral vagal trunks associated with the oesophagus.

The dorsal and ventral vagal trunks continue through the diaphragm and maintain the same relative positions at the cardia.

Dorsal Vagal Trunk

The branches coming out from the dorsal trunk mainly supply the different parts of the rumen (Figure 2.1).

Ventral Vagal Trunk

One or more branches of the ventral vagal trunk supply the left side of the cranial dorsal sac of the rumen. The majority of their branches supply the reticulum (Figure 2.2). Post-ganglionic sympathetic nerves also innervate the rumen.

The presence of myenteric plexus in the reticulum and rumen has also been recognized (Habel, 1956; Gregory, 1982).

GASTRO-OESOPHAGEAL REFLUX IN ANAESTHETIZED SHEEP

The backflow of rumen contents along the oesophagus in anaesthetized sheep will be designated as "reflux" in the text throughout.

There have been no experimental studies on gastro-oesophageal reflux (GOR) in anaesthetized sheep. Leek (1975) discusses the possible

FIGURE 2.1
Nervous innervation of the rumen (Habel, 1956).

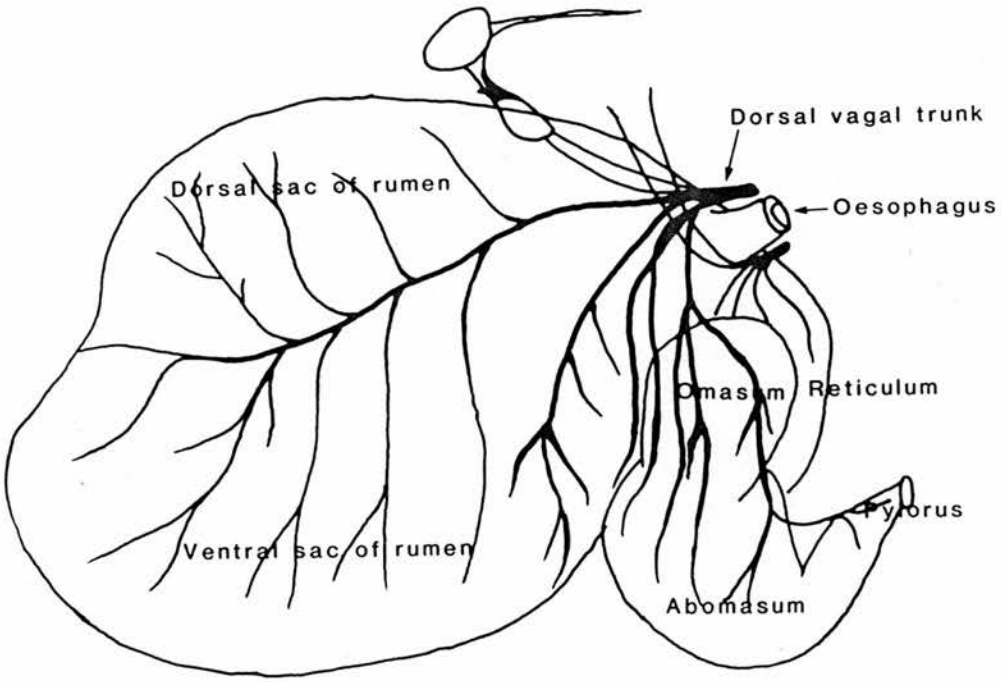
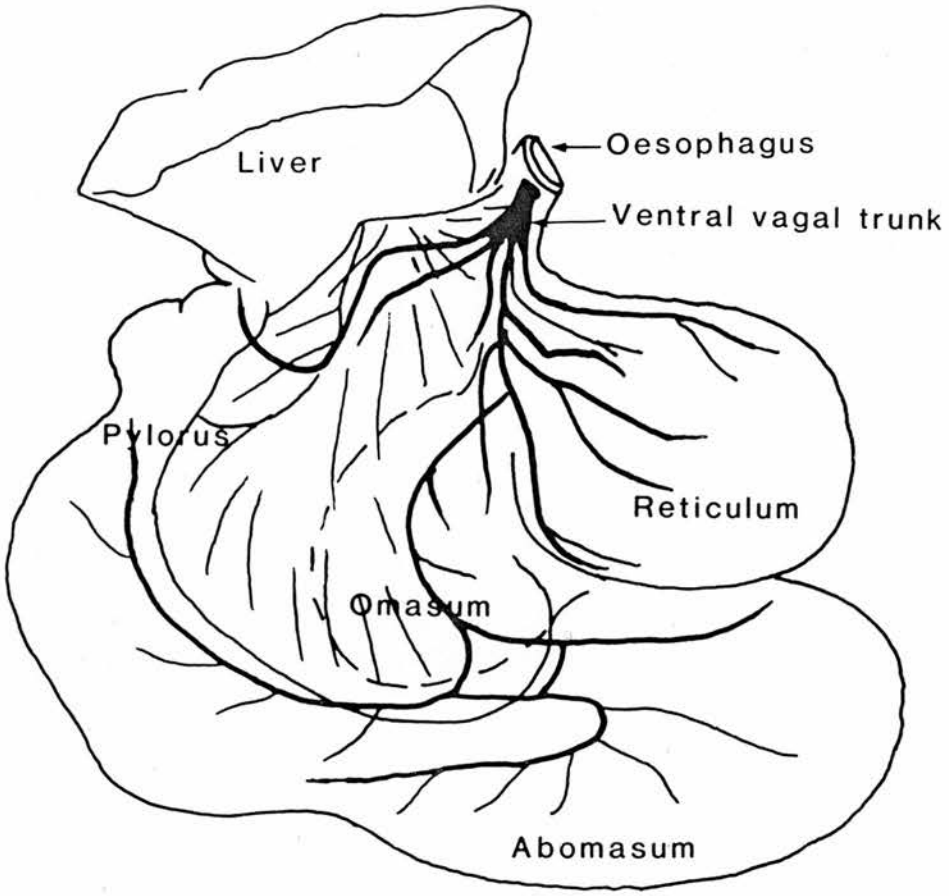


FIGURE 2.2
Nervous innervation of the reticulum (Habel, 1956).



mechanisms of GOR in ruminants. He suggests that there are four different situations when rumen contents may enter the oesophagus. These are (1) regurgitation of the cud bolus during rumination, (2) vomiting under certain experimental conditions, (3) eructation of ruminal gases, and (4) passive reflux of rumen contents through relaxed oesophageal sphincters. Leek suggests that during anaesthesia the tone of the oesophagus reduces and the cardia relaxes due to suppressive effects of anaesthetics on medullary centres. The rumen contents then pass into the relaxed oesophagus as a result of a pressure gradient across the cardia, gravitational force and bodily movements. There is no experimental evidence, however, in support of these suggestions.

EFFECT OF POSTURE ON GASTRO-OESOPHAGEAL REFLUX DURING ANAESTHESIA

Phillipson and Burnett (1939) state that gastro-oesophageal reflux in sheep may be controlled by tilting the operating table so that the posterior part of the sheep's body is about six inches lower than the anterior part. The influence of gravity on the mechanism of reflux in anaesthetized sheep has also been suggested by Leek (1975). Apart from these statements there has been no experimental work to study different postures and their possible influence on the reflux of rumen contents into the oesophagus.

EFFECT OF ANAESTHESIA ON OESOPHAGUS AND STOMACH

Little is known about the action of general anaesthetics on oesophagus and stomach. Meltzer (1899) has reported that the appearance of peristalsis in the canine oesophagus is considerably influenced by the degree of anaesthesia. He found that during deep anaesthesia

peristalsis is stopped. This degree of anaesthesia may affect the deglutitive centre by preventing the progress of the impulse along the primary afferent nerves within the centre, or prevents the transmission of its efferent impulses to the oesophagus. Botha (1959) also reports of the marked inhibition of the oesophageal peristalsis by the depth of anaesthesia.

Comline and Titchen (1951) and Duncan (1951) state that anaesthetics acting primarily on the central nervous system reduce or abolish the regular cyclic motility and reflexes of the ruminant stomach leaving only the intrinsic motility. It is reported that general anaesthetics cause cessation of co-ordinated reticuloruminal movements and these reappear when the anaesthesia is light particularly during recovery from anaesthesia (Iggo, 1956). The motility of the oesophagus is likely to be affected by the depressant action of the anaesthetics on hind brain reflexes (Leek, 1975). Leek (1975) suggests that anaesthetic agents depress medullary centres including the oesophageal centres which reduces the oesophageal tone and causes relaxation of lower oesophageal sphincter.

In the dog, the oesophageal sphincters are profoundly affected and very little or no tone is maintained in these areas during anaesthesia which removes some of the important components of the oesophageal motor actions and no high pressure is detected in the LOS area, the swallowing reflexes are also inhibited (Levitt, Dedo and Ogura, 1965; Code and Schlegel, 1968).

GASTRO-OESOPHAGEAL REFLUX IN MAN

The occurrence of gastro-oesophageal reflux (GOR) in man during anaesthesia is a problem and has been reported to be a major cause of anaesthetic related morbidity and mortality (Veter, Fell, Cotton and Smith, 1982).

Marchand (1955) offers three hypothetical causes for the occurrence of GOR. These are (1) increase of the intragastric pressure above levels normally withstood by the resisting mechanism present at the cardia, (2) loss of the normal oesophago-gastric angle, and (3) inhibition of the tone of the terminal oesophagus.

Creamer (1955) reports that the positive pressure waves resulting from the reflux of gastric contents into the thoracic oesophagus are sometimes limited to the lower oesophagus. These pressures are mainly due to entry of gastric contents and not due to retrograde peristalsis. He suggests that the entrance of gastric fluid into the oesophagus often produces peristaltic movements and that reflux is always associated with inspiration.

REGURGITATION IN SHEEP AND OTHER RUMINANTS

Regurgitation is a physiological phenomenon in which a portion of rumen contents are carried to the mouth for remastication.

Mechanism

Daubenton (1770) was the first to study the act of regurgitation (cited by Bergman and Dukes, 1926). He examined the reticular contraction in sheep at necropsy and concluded that the bolus for regurgitation is moulded by the reticulum and is forced into the cardiac end

of the oesophagus via the oesophageal groove. Fluorens (1833) disproved this idea and demonstrated that regurgitation is not inhibited by surgical fixation of the reticulum to the abdominal wall (cited by Stevens and Sellers, 1968). He added that the oesophageal groove was the bolus-forming organ in conjunction with the closed reticulo-omasal and cardiac orifices (cited by Bergman and Dukes, 1926); but Collins (1884) showed that the lips of the reticular groove were not involved in regurgitation (see Bergman and Dukes, 1926). He sutured together the lips of the groove in two bulls using wire sutures and reported that regurgitation was perfectly normal. It was suggested that during regurgitation, the rumen contents were pushed into the cardia by the combined contraction of the rumen and reticulum assisted by the fixed diaphragm and by the contraction of the abdominal muscles.

Toussant (1875) first demonstrated that the glottis remains closed during regurgitation (cited by Bergman and Dukes, 1926). He also stated that regurgitation in sheep and oxen is brought about by a special aspiratory act of the thorax and that at the moment of regurgitation, the rumen is quiescent.

It is reported that rumen, reticulum and abdominal muscles are not involved at the moment of regurgitation (Bergman and Dukes, 1926). The level of rumen contents is essential to be on or above the level of the cardia for a successful regurgitation which is also influenced by the water content of the rumen. This view was supported later by Downie (1954) who stated that the level of ingesta should be sufficiently raised for the act of regurgitation in cattle. This increased level of ingesta is contributed by the contraction of the reticulorumen.

Stigler (1931) studied the mechanism of regurgitation in goats by X-ray investigation and concluded that the hydrostatic pressure of the rumen contents is sufficient to fill the cardiac portion of the oesophagus but the pressure gradient is not sufficient to produce regurgitation (cited by Downie, 1954). X-ray observations also indicate that the ingesta are not conveyed to the oesophagus by contractions of the stomach but by a sudden inspiratory effort which produced a low pressure followed immediately by an expiratory effort and high pressure. The first phase of the action sucks the ingesta while the second phase aids the propulsion of the fluid mass forwards.

The majority of workers have suggested that a pressure gradient across the lower oesophageal sphincter (LOS) is consistently found during regurgitation (Bergman and Dukes, 1926; Dziuk and Sellers, 1955; Bell, 1958; Dougherty, 1961; Winship et al., 1964; Stevens and Sellers, 1968; Phillipson, 1970). These workers believe that the difference of pressure is produced as a result of inspiration against a closed glottis. Webster (1926) however offered a theory that shortening of the longitudinal muscles and relaxation of the circular muscles bring about such pressure difference (cited by Bell, 1961). No particular phase of respiration is associated with the act of regurgitation in most instances, but a deep inspiratory effort occurs in 25% of cases (Winship et al., 1964).

Webster and Cresswell (1957) using conscious sheep found that an intra-thoracic negative pressure is not a prerequisite for the act of regurgitation. They suggested that the negative pressure in the thoracic oesophagus is purely incidental and aspiration of reticular contents results from active dilatation of the whole of the thoracic

oesophagus. It is hypothesized that the thin-walled and easily dilatable thoracic oesophagus normally suspends loosely in the mediastinum with its walls either contracted or collapsed (Webster and Cresswell, 1957). At the moment of regurgitation, the LOS and the lower oesophagus relax and simultaneously the oesophagus increases in tension by a sudden forcible contraction of the diaphragm which produces a momentary ballooning of the oesophagus. This creates the negative pressure within the terminal portion and causes the rumen contents to siphone rapidly forwards.

Bell (1958) supported the evidence of Webster and Cresswell (1957) with the demonstration that goats fitted with tracheal cannulae regurgitate normally, although in this condition, the intra-oesophageal pressure is markedly reduced. Rumination is also observed in animals with transthoracic bilateral phrenic section with resulting diaphragmatic paralysis.

The intraruminal pressure is found to vary considerably during regurgitation, the maximum being 14.7 mm Hg, but the maximum reticular pressure is recorded as 47.7 mm Hg which falls rapidly to a resting level near atmospheric pressure (Bell, 1958). Bell (1958) also stated that the synchronous occurrence of regurgitation with inspiration and maximal contraction of the reticulum provides a differential pressure between the reticulum and oesophagus of some 44.1-58.8 mm Hg. But Winship et al. (1964) found a lower rumeno-oesophageal pressure difference of 3.0-8.0 mm Hg in sheep, goats and cattle. Dougherty and Habel (1955) showed in sheep that complete filling of rumen and reticulum with hot water at a pressure of 40.0 mm Hg does not produce regurgitation.

The majority of investigations demonstrate that rumen contents during regurgitation are carried to the oro-pharynx by active oesophageal antiperistalsis (Bell, 1958; Dougherty, 1961; Winship et al., 1964; Stevens and Sellers, 1968; Flaih and Singleton, 1978). The velocity of antiperistalsis during rumination in sheep is 53.8 cm/sec (Winship et al., 1964). The electromyographic study of Dougherty, Hill, Cook and Riley (1971) suggested that during rumination, antiperistalsis is at a mean rate of 182 cm/sec. Flaih and Singleton (1978) calculated a velocity of 86 cm/sec.

The mechanism of regurgitation in the conscious ruminant is described by Stevens and Sellers (1968). A contraction of the reticulum floods the cardia with ingesta. An aspiratory effort of the thorax then occurs simultaneously with the submersion of the cardia and with dilatation of the caudal oesophageal sphincter. As a consequence, ingesta enters the thoracic oesophagus. An antiperistaltic oesophageal contraction then carries the refluxed materials to the oro-pharynx.

The electromyographic study of Titchen (1979) demonstrated in sheep that the costal but not the hiatal fibres of the diaphragm are involved at the time of regurgitation.

Radiologically, regurgitation in sheep appears to be associated with an extra reticular contraction followed by normal double contraction (Phillipson, 1970; Deswysen and Ehrlein, 1979). This extra contraction aids the course material to move from the upper part of the reticulum into the cranial sac of the rumen and the contents of the lower part of the reticulum are carried to the LOS for regurgitation (Deswysen and Ehrlein, 1979; Ehrlein, 1979).

The Role of LOS in Regurgitation

Kay (1983) stated that regurgitation during rumination occurs during an extra contraction of the rumen. The cardia relaxes and aspiration of digesta into the lower oesophagus is achieved by simultaneous contraction of the abdominal muscles and reduction of thoracic pressure by an inspiratory movement with the glottis closed. Antiperistalsis then carries the bolus to the oro-pharynx.

Different workers have given different views about the contribution of gastric and oesophageal movements in the act of regurgitation - but the relaxation of LOS has been demonstrated by many investigators (Bell, 1958; 1961; Stevens and Sellers, 1960; 1968).

The role of LOS in regulating the act of regurgitation may be essential (Laitinen, Mokka, Valanne and Larmi, 1978); but Winship et al. (1964) failed to detect any evidence for such sphincter in sheep by using a pull-through method with open tip catheters. They found no high pressure zone at the level of gastro-oesophageal junction.

Titchen (1979) reports in sheep that contraction of the caudal thoracic oesophagus develops over a period of 2 sec before and ceases at the time of more forceful inspiratory effort associated with regurgitation. He states that the contraction of caudal thoracic oesophagus within 1.5-2.5 cm of the hiatus oesophagus is characteristic of LOS.

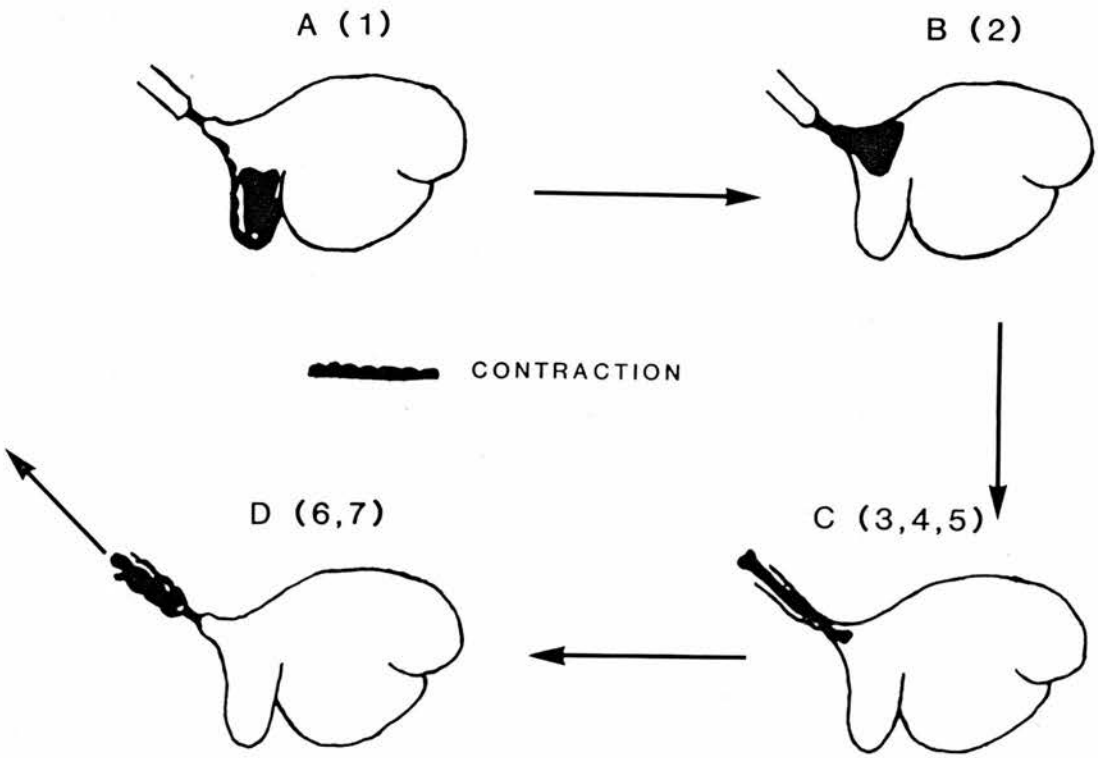
From the above information, the sequence of regurgitation may be diagrammatically represented as shown in Figure 2.3.

Control of Regurgitation

In sheep, regurgitation is normally dependent on the integrity of vagus nerve since total vagotomy abolishes rumination (Duncan, 1953; Habel, 1956).

FIGURE 2.3

Sequence of Regurgitation in Conscious Ruminant



1. EXTRA RETICULAR CONTRACTION
2. FLOODING OF THE LOWER OESOPHAGEAL SPHINCTER (LOS) WITH INGESTA
3. PRESSURE GRADIENT ACROSS THE LOS
4. RELAXATION OF THE LOS
5. ENTRANCE OF RUMEN CONTENTS INTO THE OESOPHAGUS
6. ANTIPERISTALSIS OF THE OESOPHAGUS
7. GLOTTIS CLOSED AND SOFT PALATE RAISED

The early report by Clark (1953) that the controlling centre for reticuloruminal motility was located in the sub-cortical area anterior to the pituitory infundibulum found no support later when the centre was demonstrated in the medulla oblongata (Bell and Lawn, 1955; Habel, 1956; Bell, 1961; Dougherty, 1961).

Stevens and Sellers (1959) found that the afferent fibres for the regurgitation reflex are present in the ventral abdominal vagus. These afferents are possibly stimulated by the presence of coarse food materials in the rumen and reticulum (Dougherty, 1961).

ERUCTATION IN SHEEP AND OTHER RUMINANTS

Eructation is a physiological event where the fermented gases from the reticulorumen are expelled through the mouth. It involves the co-ordinated motor action of reticulorumen and the whole of the oesophagus. Sometimes there is some liquid gastro-oesophageal reflux associated with eructation. It is reported by Kölling (1975) that a healthy sheep in the conscious state eructates about 175 litres of gases in 24 hours and the average number of eructation is 756.

The mechanism of eructation is to some extent similar to regurgitation e.g. both are associated with reticuloruminal contractions, LOS relaxation and oesophageal antiperistalsis.

Mechanism: Conscious and Decerebrate Preparation

Eructation is a co-ordinated reflex mechanism involving rumen, reticulum, LOS and oesophagus (Clerk, 1950; Weiss, 1953; Fioramonti, Bueno, Ooms and Ruckebusch, 1982). It is suggested that the reflex phenomena in sheep and cattle associated with eructation are: a forward

moving contraction of the rumen; opening of LOS by the contraction of lateral and medial pillars of the reticuloruminal fold and the clearing of the LOS area of fluid ingesta by reticular relaxation. This effort is supplemented by strong contractions of the reticuloruminal fold, the anterior pillar and the surrounding ruminal and reticular wall (Dougherty, 1961).

Dougherty and Habel (1955) found that the motor events associated with eructation are two contractions of the reticulum, contraction and raising of the reticuloruminal fold, general ruminal tonus, relaxation of the caudal oesophageal sphincters, and gaseous distension of the oesophagus.

Many workers have reported that the gas is expelled by rapid oesophageal antiperistalsis (Dougherty, 1961; Dougherty et al., 1971; Ali and Singleton, 1974; Swenson, 1977; Flaih and Singleton, 1978). This velocity of antiperistalsis in sheep varies from 151-200 cm/sec. In bovines, this velocity is 227 cm/sec (Stevens and Sellers, 1960).

Eructation is usually associated with the secondary contraction of the dorsal rumen sac (Nichols, 1951; Williams, 1955; Stevens and Sellers, 1959; Titchen, 1968) and about one-third of the total number of eructations occur in conjunction with a primary ruminal contraction. Ali and Singleton (1974) however report that in sheep, the spontaneous eructations are more often associated with primary contractions than with secondary. The rate of eructation varies with the rate and amplitude of rumen contractions even when the cardia is continuously exposed to gas below. It has been reported that the motility of the rumen and reticulum is not essential for eructation (Dougherty and Meredith, 1955; Dougherty, 1961).

Carr, Scot and Titchen (1979) have studied the oesophageal reactions during eructation in sheep by manometry and electromyography and state that during eructation there is a biphasic increase in pressure with an initial small but sustained increase in pressure. This occurs first in the caudal thoracic oesophagus and later in the caudal cervical oesophagus and is followed by swallowing. The reticulum remains inactive at the time of eructation and the movement of the costal and hiatal fibres of the diaphragm ceases. It is suggested that the degree of oesophageal contraction reflects the afferent stimulation arising during eructation. Oesophageal contraction involves a variable degree of active oesophageal activity and the passive movement of gas into the oesophagus contributes significantly in eructation. It is concluded that during eructation oesophageal contractions may facilitate the movement of gas orally in the oesophagus but are not essential for its occurrence.

Many workers have reported the sequence of eructation in ruminants (Williams, 1955; Ali and Singleton, 1974; Swenson, 1977). According to Williams (1955) these are relaxation of the reticulum, passing of the ruminal gases into the cardia region by the extraruminal contraction, raising and opening of the LOS by the corresponding contraction of the transverse fold and the surrounding rumen wall and movement of gas into the caudal thoracic oesophagus which initiates the eructation process.

Ali and Singleton (1974) have found that in the beginning of eructation there is a fall in pressure in the lower part of the cervical oesophagus and occasionally in the thoracic oesophagus followed by an antiperistaltic wave that commences at the cardia and travels only to the levels of the first rib and is never extended into the cervical oesophagus.

According to Swenson (1977) the first event during eructation is the reflex relaxation of the LOS following the excitation of the distension sensitive receptors in the rumen wall. The gas then enters the oesophagus through the relaxed cardia aided by rumen contraction and an abdominal pressure and is then expelled by antiperistalsis. Eructation is normally followed by swallowing. The spontaneous eructations are usually associated with rumen contraction but those resulting from intraruminal insufflation are not associated with rumen contraction.

The sequence of eructation in conscious ruminants may be summarised diagrammatically as represented in Figure 2.4.

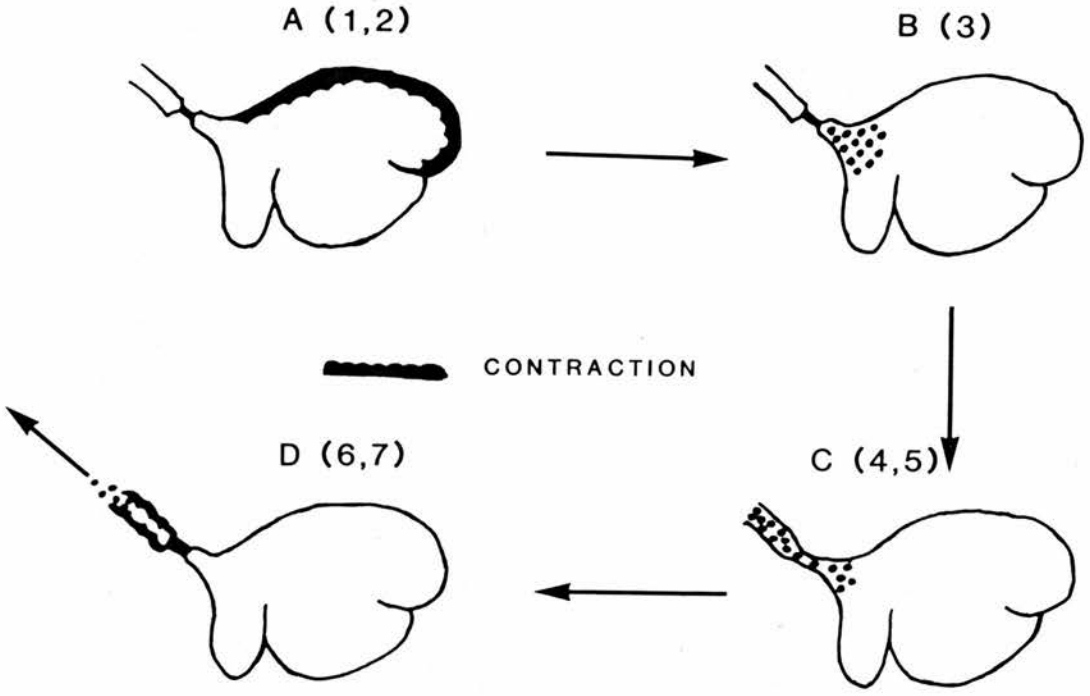
Initiation of Eructation

Mead, Cole and Regan (1944) suggested that coarse fibres of the diet elicit the eructation reflex in cattle. Weiss (1953) however stated that the presence or absence of coarse roughage in the rumen does not appear to affect the eructation reflex nor apparently is it the stimulus for its initiation. He suggested that intraruminal pressure by gas is the main stimulus for the eructation reflex. This observation has been supported by other workers (Dougherty et al., 1958; Dougherty, 1961; Reid and Titchen, 1965). They were able to demonstrate in decerebrate sheep an increased rate of eructation following insufflation of the rumen and reticulum with gas. It is also reported that eructation is more likely to occur in sternal position than lateral recumbancy.

Moderate electrical stimuli applied to the intact thoracic dorsal vagal trunk in cattle elicits an increased rate of rumen contraction

FIGURE 2.4

Sequence of Eructation in Conscious Ruminant



1. SECONDARY CONTRACTION OF THE DORSAL RUMEN SAC
2. CLEARING UP OF THE LOWER OESOPHAGEAL SPHINCTER (LOS)
3. DISTENTION OF THE LOS AREA WITH GAS
4. RELAXATION OF THE LOS
5. ENTRANCE OF GAS INTO THE OESOPHAGUS
6. RAPID ANTIPERISTALSIS OF THE OESOPHAGUS
7. GLOTTIS REMAINS OPEN

(Dziuk and Sellers, 1955). The increased frequency of reticuloruminal motility is associated with an increased rate of eructation. The increased intraruminal pressure causes increased eructation. Cholinergic drugs such as pilocarpine increase the frequency of secondary contractions of the rumen, but the LOS pressure is reinforced and less eructation occurs (Fioramonti et al., 1982).

Inhibition of Eructation

Eructation may be inhibited by different factors. Weiss (1953) reported in sheep, goats and cattle that eructation is prevented by mechanical obstruction in the oesophagus, frothing of the ingesta, the degree of filling of the rumen and posture, elevation of hind quarters, distension of the abomasum or caecum and atropinization. Filling of the rumen and elevation of hind quarters impede eructation by increasing the distance between free gas and cardia. The distension of abomasum or caecum with either fluid or air retards eructation through reflex inhibition of reticulorumen motility.

Inhibition of eructation also occurs when the cardia is flooded with water, ingesta or mineral oil (Dougherty and Meredith, 1955; Dougherty et al., 1958; Dougherty, 1961; Dougherty, Mullenax and Allison, 1965). In the decerebrate preparation in dorsal recumbancy and with a well-filled rumen, eructation is completely inhibited even when the rumen is insufflated to raise the pressure up to 30.0 mm Hg (Dougherty et al, 1958). They also reported that with an empty rumen, the decerebrate sheep eructates as efficiently in dorsal recumbancy as in normal standing position (Dougherty et al., 1958).

Nichols (1951) reported that in conscious sheep overloading of the rumen with water coupled with gaseous insufflation progressively inhibits

eructation but with further insufflation, the animal died fairly rapidly due to cessation of breathing and consciousness.

Stevens and Sellers (1956) found that injection of procaine hydrochloride solution on the dorsal vagal trunk in cattle impairs eructation efficiency with a rise in intraruminal pressure. Electrical stimulation posterior to the procaine block is ineffective but when applied anterior to the procaine block causes increased rate of eructation. Later, in 1959, they demonstrated that procainization of the cardiac mucosa appears to allow a rapid eructation apart from rumen contraction suggesting that this sphincter area is an important effector organ in the eructation reflex. Atropinization reduces eructation rate at atmospheric intraruminal pressure and during insufflation (Stevens and Sellers, 1959).

Section of the right ventral branch of the vagus causes abomasal distension and results in inefficient eructation due to inhibition of reticular activity (Weiss, 1953). Weiss (1953) also reported that section of the left dorsal branch diminishes the strength of ruminal contractions and eructation efficiency.

Control of Eructation

The controlling centre for eructation may be located in the brain stem (Dougherty, 1961). The reflex responsible for eructation involves integration in the vagus nerve since total vagotomy abolishes reticuloruminal contractions and inhibits eructation (Habel, 1956). Stevens and Sellers (1959) stated that the afferent fibres for the eructation reflex are present in the dorsal abdominal vagus.

ERUCTATION IN MAN

The pressure studies of Creamer (1955) utilizing fluid-filled polyethylene catheters with Hansen Capacitance manometers demonstrated that there is no associated antiperistalsis at the time of eructation and a positive pressure occurs simultaneously in all parts of the oesophagus during the event. The radiographic and manometric investigation by McNally, Kelly and Ingelfinger (1964) showed that reflex relaxation of LOS is responsible for eructation. On insufflation, the intra-oesophageal pressures become identical to that of gastric pressures. They found no correlation between the intragastric pressure and eructation frequency.

UPPER OESOPHAGEAL SPHINCTER

The upper oesophageal sphincter (UOS) is defined as an intraluminal zone of high pressure that exists between the pharynx and the oesophageal body (Goyal and Cobb, 1981). The UOS remains tonically closed under resting conditions (Botha, 1959) over a distance of 1.0-3.0 cm by a band of striated muscle composed of cricopharyngeal and probably also of oesophageal elements (Ingelfinger, 1958). The main reason for this tonic closure may be to avoid air being sucked into the oesophagus on inspiration (Ingelfinger, 1958). This tonic closure of UOS also prevents aspiration of ingesta from the oesophagus into the lungs and therefore it acts as a second closing mechanism of the stomach. During anaesthesia, this zone may offer resistance to the flow of refluxed rumen contents and prevent reflux into the pharynx.

Anatomically, the UOS is formed by the overlapping of pharyngeal and oesophageal musculature (Sisson and Grossman, 1975). Physiologically,

it maintains a continuous state of tone or pressure due to contraction of its muscle fibres (Schlegel and Code, 1958). This tone occurs in response to a continuous impulse activity in its efferent nerves (Schlegel and Code, 1958). During swallowing, this impulse activity is interrupted briefly and the sphincter relaxes. The relaxation of the sphincter at the initiation of deglutition is also reported by Code, Creamer, Schlegel, Olsen, Donoghue and Anderson (1958), Pope (1970), Lipshutz and Cohen (1971).

There are three possible mechanisms of sphincteric opening: (1) the sphincter opens passively in response to oro-pharyngeal pressure, (2) it is pulled apart by other muscles or (3) its intrinsic tonicity is inhibited (Ingelfinger, 1958).

The evidence for active closure of this sphincter is still controversial (Roman and Gonella, 1981) because of the absence of tonic EMG activity in the muscles surrounding the pharyngo-oesophageal junction specially the cricopharyngeus. Doty (1968) concluded on this issue that the sphincter remains closed by passive elasticity of surrounding tissues, that active closure is readily induced by a variety of responses serving to contract the cricopharyngeus and that during swallowing the orifice is opened passively by hyoid and laryngeal movements while all contraction of cricopharyngeus is initially forestalled by the swallowing centre exerting a powerful inhibition on cricopharyngeal motoneurons. It is stated however that at rest the cricopharyngeus exhibits a continuous spiking activity which with a swallow is completely inhibited while intraluminal pressure falls. This inhibition is followed by an intense burst of spikes which corresponds to the deglutitive contraction of the pharyngo-oesophageal sphincter and then

the EMG activity and the intraluminal pressure return to their resting level. The mean resting pressure of the sphincter in dogs is 14.7 mm Hg (Schlegel and Code, 1958).

DEGLUTITION AND ITS CONTROL

Deglutition or swallowing is a voluntary and involuntary act by which food is taken through the mouth, pharynx and upper oesophageal sphincter into the cranial cervical oesophagus. In a light plane of anaesthesia swallowing may also occur and may influence the occurrence of gastro-oesophageal reflux. A brief review of the mechanism is therefore recorded.

Deglutition is a reflex which once initiated voluntarily thereafter becomes involuntary and as the bolus moves backward, sensory receptors are activated which initiate the involuntary phase of the deglutition reflex (Botha, 1962b; Code and Schlegel, 1968; Goyal and Cobb, 1981). During deglutition enteroceptors are stimulated in the mucous membrane of the sensory areas over the base of the tongue, epiglottis, soft palate and retro-pharyngeal wall, when afferent impulses are set up and carried to the higher centres by the glossopharyngeal nerve and laryngeal branches of the vagus (Botha, 1962b). From the higher and medullary centres, motor impulses are carried in the ninth, tenth and eleventh cranial nerves to the entire length of the oesophagus to initiate reflex deglutition.

The entrance of food material into the pharynx is due to the pressure gradient - a negative suction force from below together with a positive force from above (Botha, 1962a).

The initial portion of the act of deglutition has been reported (Rushmer and Hendron, 1951; Atkinson, Kramer, Wyman and Ingelfinger, 1957; Sokol, Heitmann, Wolf and Cohen, 1966). According to these workers, the mouth is first closed, then the passage between the mouth and the nose, and finally the entry way to the lungs. The mouth is closed by the lips; connection to the nasal cavities is closed by simultaneous contraction of the levator palati and palato-pharyngeal muscle, which raise the soft palate and approximate the posterior pillars; closure of the connection to the respiratory pathway is by elevation of the larynx with closure of the glottis while respiration is momentarily inhibited. After these initial activities, the caudad transport of the bolus is accomplished by orderly sequential contraction of the muscles of the tongue and the pharynx with co-ordinated relaxation and subsequent contraction of the pharyngo-oesophageal sphincter (Code and Schlegel, 1968). The peristaltic sequence of contraction produces an increase of pressure which starts at the upper surface of the tongue and sweeps continually caudad through the mouth and pharynx into the pharyngo-oesophageal sphincter and is then propelled through the sphincter into the oesophagus at the speed of 40-50 cm/sec.

Fyke and Code (1955) demonstrated in man that with the initiation of the act of swallowing, a high positive pressure ($\bar{x}$ 74.0 mm Hg) suddenly develops in the upper part of the pharynx. Simultaneously, a negative pressure occurs at the pharyngo-oesophageal junction and in the upper portion of the oesophagus as an integral part of the swallowing reflex which facilitates the transport of material through this region. The high pressure which develops in the upper part of

the pharynx at the onset of swallowing, sweeps as a wave through the junctional region into the upper portion of the oesophagus where it becomes continuous with the primary peristaltic wave (Fyke and Code, 1955).

The food bolus after swallowing is carried down to the LOS by oesophageal peristalsis (Weisbrodt, 1974). The LOS then relaxes and allows the bolus to enter the stomach and then again contracts. The peristaltic wave is not essential when the bolus is liquid which can go down by the influence of gravity. It is concluded that the relaxation of LOS during swallowing occurs as an integral part of the deglutition reflex (Weisbrodt, 1974). Pert, Davidson, Almy and Sleisenger (1959) suggested that in man this relaxation may be due to relaxation of tonically contracted and circularly placed smooth muscle fibres. The LOS then closes with a prolonged contraction as the peristaltic wave passes into it.

Summary

Deglutition is a reflex mechanism which is initiated by the voluntary intake of food. After the initiation of this reflex by the presence of food material in the pharynx, the process becomes entirely involuntary. Afferent impulses are initiated by the stimulation of enteroceptors and are carried to the higher centres through the vagus while the motor impulses are sent to the entire oesophagus to initiate or modulate oesophageal peristalsis.

VOMITING

This is the forcible expulsion of gastric contents through the oesophagus and pharynx. Vomiting usually occurs in man and other monogastric animals. In ruminants, the occurrence is unusual in its truest form because of the remote position of the abomasum from the oesophagus. The act of vomiting in the ruminant however, involves a mechanism which may be comparable with regurgitation or reflux of rumen contents.

Mongs, Salducci and Naudy (1976) recorded the diaphragmatic EMG in conscious dogs during induced vomiting and demonstrated that during retching, the diaphragmatic electrodes record a large burst of spike potentials simultaneously. No activity during the expulsive phase is recorded by the electrodes of the hiatal margin fibres, while a large and sustained burst of spike potential is recorded by the electrodes fixed in the dome of the diaphragm. They concluded that during vomiting, the diaphragmatic dome fibres are in a contracted state while muscle fibres surrounding the oesophageal hiatus are in a state of relaxation allowing the passage of gastric contents into the thoracic oesophagus.

The sequence of vomiting has been studied in unanaesthetized dogs by chronically implanted electrodes (Mongs, Salducci and Naudy, 1978). They recorded the EMG during retching and expulsive phases of vomiting. The retching phase was characterised by a series of violent rhythmic abdomino-thoracic contractions and during the expulsive phase, the animal thrusts its head forward, opens its mouth wide and contracts its abdominal and diaphragmatic muscles throughout the forceful ejection of gastric material through the mouth. During retching, the upper

oesophageal sphincter remains completely relaxed and during the expulsive phase the sphincter remains widely open.

Marchand (1955) regarded vomiting as a highly complex reflex involving spasmodic contraction of the muscles of expiration, particularly of the diaphragm and abdominal musculature. During vomiting, the oesophagus plays a passive role and does not show any antiperistalsis (Hatcher, 1924; Borison and Wang, 1953).

INITIATION AND CONTROL OF OESOPHAGEAL PERISTALSIS

Like most parts of the alimentary canal, the oesophagus undergoes peristalsis. The oesophageal peristalsis is an aboral wave-like progression of alternate relaxation and contraction of involuntary muscle. This activity of the oesophagus was first investigated in the anaesthetized dog by Meltzer (1899) who reported that oesophageal peristalsis is caused by intrinsic reflexes, i.e. from each part of the oesophagus itself and that peristalsis does not occur in the ligated oesophagus. Meltzer and Auer (1906) injected fluid and air into the oesophagus of a rabbit and observed that a peristaltic wave proceeds down the oesophagus and terminates in a contraction of the cardia. They also showed that this wave, unlike the peristaltic wave of deglutition, depends on continuous sensory impulses produced by the distending bolus for its progress down the oesophagus. This original research of studying the oesophageal peristaltic activities afterwards stimulated many workers to investigate the control mechanism of oesophageal peristalsis in various species.

There are two types of oesophageal peristalsis: primary and secondary. The primary peristalsis is initiated from deglutition by

the stimulation of the vagal afferent fibres (Cohen, Kravitz and Snape, 1978). The secondary oesophageal peristalsis is defined as the oesophageal response without the oro-pharyngeal component which is seen with oesophageal distension (Goyal and Cobb, 1981). Secondary peristalsis is independent of swallowing and is entirely involuntary (Botha, 1962a). In the striated muscle portion, secondary peristalsis is initiated by the serial discharge of efferent impulses from the medullary centres which are reflexly stimulated by afferent impulses from stretch receptors in the oesophageal wall. Botha (1962a) stated that this type of peristalsis is dependent on local reflexes in the smooth muscle portion but is also influenced by the medullary centres. It is similar to primary contraction, travels at the same speed and is dependent on the same extrinsic and intrinsic nerve supply for a normal co-ordinated completion. Secondary peristalsis usually occurs when the oesophagus is insufficiently emptied, when it is obstructed or distended or when there is reflux or regurgitation of gastric contents into the oesophagus (Botha, 1962a). The control mechanism of secondary peristalsis in man is regulated both by intraluminal volume and pH (Corazziari, Pozzessere, Dani, Anzini and Torsoli, 1978).

In man oesophageal peristalsis can be initiated by mechanical distension of the lower oesophagus (Dornhorst, Harrison and Pierce, 1954) and Mukhopadhyay and Weisbrodt (1975) demonstrated in the opossum that electrical excitation of the distal cut end of the cervical vagus initiated peristalsis. This indicates that oesophageal peristalsis is at least partly controlled by a local mechanism within the oesophagus and not extrinsically by the central nervous system. It is

suggested that the propagative nature of oesophageal peristalsis may be due to a local neuro-muscular apparatus which responds to central neural or artificially applied stimuli reaching the oesophageal wall. The authors interpreted their findings as suggesting there is the involvement of the myenteric plexus in initiating and sustaining oesophageal peristalsis. This evidence is supported by Weisbrodt (1974) who reported that oesophageal peristalsis is maintained by the central nervous system, peripheral nervous system and oesophageal muscle.

The in vitro study of Mann, Code, Schlegel and Ellis (1968) on guinea-pig and kitten oesophagi suggested that the lower oesophagus and its sphincter can function in a relatively normal fashion when completely isolated and that distension of lower oesophagus produces responses in the sphincter that are transmitted to the sphincter via the wall of the oesophagus, completely independently of the external nerve supply for both smooth and skeletal muscle.

Surna, Daniel and Waterfall (1978) stated that oesophageal contractions and LOS relaxations following electrical excitation (20-40 V, 100-400 ms) are not blocked by atropine (100 $\mu g\ kg^{-1}$ i.v.), hexamethonium (10.0 $mg\ kg^{-1}$ i.v.) or tetrodotoxin (20-40 μg intra-arterially) in anaesthetized opossums. Also, propagated contractions due to induced swallows and accompanying LOS relaxation are not blocked by any of the above drugs. These findings support the myogenic control of the oesophagus. Christensen and Robinson (1982) however reported that the myenteric plexus is more dense in the oesophageal body than its sphincters in the opossum which suggests that oesophageal peristalsis is more neurogenic in control. Falempin, Madhloum and Rousseau (1983) denervated the afferent fibres of the thoracic oesophagus in

anaesthetized sheep at the level of nodose ganglion and demonstrated that vagal input is essential to initiate primary oesophageal contractions resulting from swallowing.

Ingelfinger (1958) hypothesized that certain centres in the brain send motor impulses to progressively distal portions of the oesophagus giving rise to co-ordinated peristaltic contractions. This concept was later supported by the finding that bilateral vagotomy reduces the oesophageal motor response to pharyngeal stimulation in the opossum (Mukhopadhyay and Weisbrodt, 1975).

Roman and Gonella (1981) reported that during primary oesophageal peristalsis resulting from the swallowing of a bolus, vagal motor discharge is more powerful than the discharge produced during secondary peristalsis.

Ingelfinger (1958) described the entire peristaltic phenomenon in man as follows: A swallow activates a medullary swallowing centre which releases inhibitory and excitatory phenomena controlling the oro-pharyngeal motor sequence of swallowing. Under the influence of the inhibitory phase, the upper oesophageal sphincter relaxes 0.2-0.3 sec after the beginning of swallowing. Then the wave of inhibition sweeps down the rest of the oesophagus to reach the LOS in 1.5-2.5 sec after swallowing. To affect oesophageal muscle, the inhibitory wave requires intact vagi and myenteric ganglia. After swallowing, when the peristaltic wave of pharyngeal constriction reaches the upper oesophageal sphincter, it closes, usually in excess of resting tonus and is followed by a wave of contraction along the oesophagus and as soon as the peristaltic wave arrives at the LOS, the sphincter undergoes a strong and prolonged after-contraction. The distension of the oesophagus without

any swallowing elicits a similar sequence of inhibition-contraction to move over the oesophagus and the LOS which suggests that the propagation of both phases of peristalsis requires normal myenteric and vagal functions (Ingelfinger, 1958).

Creamer and Schlegel (1957) commented from their study in man that the motor control of the oesophagus is composed of many discrete reflexes and all these are involved in the act of swallowing.

The central mechanism responsible for oesophageal motor activities is located in the deglutition centre in the medulla (Roman and Gonella, 1981). The motor sequence of deglutition depends on a central mechanism which, after starting, could run its entire length without any further afferent activity. In sheep, in the absence of afferent input, the central mechanism is able to organise the motor sequence of peristalsis throughout the oesophagus.

Stimulation of pharyngo-oesophageal nerves in the dog and cat caused strong contraction of the cervical portion of the oesophagus (Hwang, Grossman and Ivy, 1948). In chronic experiments in these species, bilateral section of the vagal trunk results in paralysis of the oesophagus which is also found in sheep (Newhook and Titchen, 1976).

Christensen and Lund (1969) report that peristalsis can occur in an electrically or mechanically stimulated oesophagus placed in an isolated organ bath.

In anaesthetized opossums bilateral vagotomy significantly decreases the primary oesophageal peristalsis to a pharyngeal stimulation but does not reduce or eliminate secondary peristalsis to a balloon distension (Ryan, Snape and Cohen, 1977). It was concluded that vagus

nerve is of primary importance in regulating the oesophageal response to pharyngeal stimulation and that local neuromuscular factors within the oesophageal wall appear sufficient to maintain an oesophageal response to distension in this species.

In humans, distension of the oesophagus with a balloon often causes peristalsis (Creamer and Schlegel, 1957; Code et al., 1958). The relaxation of the LOS persists until the perstaltic wave reaches it. It has been suggested that the cervical oesophagus is the most sensitive, the distal oesophagus less sensitive and the middle part least sensitive to balloon distension. Repeated contractions are observed above the distension while there is an inhibition distal to it (Creamer and Schlegel, 1957). Winship et al. (1964) however, reported in sheep that distension of balloon in the distal one-third of the oesophagus initiates secondary peristalsis distal to the balloon but never proximal to the distending stimulus.

In decerebrate sheep, moderate distension of a balloon in the oesophagus evokes a series of contractions which are not accompanied by bucco-pharyngeal or upper cervical oesophageal movements of swallowing (Sellers and Titchen, 1959). Distension of a balloon in the reticulum increases the frequency of caudal thoracic oesophageal contractions suggesting that the activity of oesophagus in ruminants may be modified by conditions or activity in the stomach.

Creamer (1955) reported in man that when gastric fluid appears in the oesophagus, a propulsive wave is often reflexly initiated which progresses normally to the cardia. This wave seems to be caused by the presence of fluid in the oesophagus.

Summary

The initiation and control of the primary and secondary types of oesophageal peristalsis are different. Primary oesophageal peristalsis is initiated from deglutition and mediated through the vagus nerve. Secondary oesophageal peristalsis, however, is independent of swallowing and is initiated from the stimulation of vagal mechanoreceptors in the oesophageal wall. The oesophageal peristalsis seems to be controlled by the integrated activity of the intrinsic and extrinsic nervous systems and oesophageal musculature.

CHARACTERISTICS OF OESOPHAGEAL PRESSURES

Sheep

Little information is available about the characteristics of oesophageal pressure changes in sheep particularly in the anaesthetized condition. However, Winship et al. (1964) found that the duration of the peristaltic wave in the distal oesophagus is greater than in the proximal part. The velocity of the oesophageal pressure wave during swallowing is 25 cm/sec as determined by using two open tip catheters, one in the mid cervical oesophagus and the other in the thoracic oesophagus (Dougherty et al., 1971).

Other Species

In man, the duration of peristalsis to traverse the oesophagus varies from 2.6-4.0 sec, the intraluminal pressure amplitude from 20-165 mm Hg and velocity from 1.83-4.0 cm/sec (Dornhorst, Harrison and Pierce, 1954; Stef, Dodds, Hogan, Linehan and Stewart, 1974; Mukhopadhyay and Weisbrodt, 1975; Wallin, Madsen and Boesby, 1983). The mean velocity of the peristaltic wave in the upper oesophagus varies from 2.92-

3.29 cm/sec while in the lower oesophagus it increases to 4.98 cm/sec at the level of 7.5 cm above the lower oesophageal sphincter (LOS) and then falls to 2.15 cm/sec (Humphries and Castell, 1977). Mukhopadhyay and Weisbrodt (1975) however, reported that the velocity of peristalsis in the lower third of the oesophagus progressively decreases from 3.25-1.83 cm/sec. No significant change in mean intra-oesophageal pressure amplitude, however, was observed (from 53.4-69.5 mm Hg) in the lower two-thirds of the oesophagus although the amplitude of the peristaltic wave tends to be larger in the lower third of the oesophagus (Humphries and Castell, 1977).

Stef et al. (1974) found that the peristaltic complexes in man were larger in amplitude (165 mm Hg) and shorter in duration (2.6 sec) in the cervical oesophagus than in the thoracic oesophagus. In dogs, the velocity of peristalsis was faster in the upper part of the oesophagus, while in man it was faster in the lower part of the oesophagus. The high pressure zone at the gastro-oesophageal junction exists in the dog and at rest the LOS undergoes changes in tone often with sudden generation or release of large pressures (Schlegel and Code, 1958). They also reported that the patterns of behaviour of LOS are independent and such changes do not occur in the body of the oesophagus.

Lind, Warrian and Wankling (1966) found that the resting pressure within the body of the oesophagus in man was 1.1 mm Hg and the pressure at different points was uniform under resting conditions.

Code et al. (1958) showed that in the centre of the human LOS, the pressure fluctuations with breathing become reversed. The pressure towards the lower oesophagus decreases with inspiration while in the stomach, it increases. Sometimes respiratory pressures may be picked

up by the LOS tracing. The significant fluctuation of the LOS pressure as resulted from atropinization or abdominal compression does not affect the resting pressure in the oesophagus (Lind, Crispin and McIver, 1968).

ELECTRICAL ACTIVITIES OF THE OESOPHAGUS AND ITS SPHINCTERS

Electromyography (EMG) is a method used to study the electrical activities of the muscles during activity. Few reports exist of oesophageal electrical activity. Hellemans, Ventrappen, Valembois, Janssens and Vandenbrougkf (1968) found no electrical activity in the resting LOS and body of the oesophagus in the dog, cat and monkey. Smooth muscle spikes (a graphical recording of transient variation in electric action potential characterised by a sharp peak) have not been observed at rest. Spikes of low amplitude are observed occasionally in the striated muscle at rest. The electrical changes of the sphincter muscle during deglutition are a burst of spikes similar to that observed in the oesophagus. It is also reported that relaxation of LOS is not associated with any electrical change (Hellemans et al., 1968). This absence of specific resting electrical activity in the LOS indicates that there is a marked discordance between the manometric and electrical observations on the canine LOS.

Weisbrodt (1974) reported that oesophageal muscle has no basic electrical rhythm or slow waves. However, the absence of an electrogram does not necessarily exclude the occurrence of muscular contraction (Itabisashi, 1970). This view is also shared by Goyal and Gidda (1981) who reported that in opossums the EMG may not be correlated with mechanogram and that lower frequencies of stimulation may be associated

with electrical spike burst without mechanical contraction whereas higher frequencies of vagal stimulation may cause mechanical contraction without associated spike bursts.

Asoh and Goyal (1978) investigated in anaesthetized opossums the electrical activity of LOS and found that the sphincter exhibits a distinctive spike activity at rest although this spike activity is not the sole determinant of the resting sphincter pressure which may be divided into spike-independent and spike-related components. They reported that more than 55-75% of the LOS pressure is spike-independent and there is no direct correlation between the spike potential activity and the LOS pressure. The in vitro study of Daniel, Taylor and Holman (1976) in the opossum indicates that the active potential of LOS at rest is myogenic.

LOWER OESOPHAGEAL SPHINCTER (LOS)

The definition of LOS is controversial. There are reports suggesting anatomical and physiological evidences for the presence of LOS. Rinaldo, Levey, Smathers, Gardner and McGinnis (1971) reported that in the dog the LOS is characterised by muscular thickening which is about 1.0 cm long and composed of smooth muscle. Most of the investigators however, support the physiological evidence for the existence of LOS (Code et al., 1958; Schlegel and Code, 1958; Magee, 1962; Weisbrodt, 1974).

The physiological sphincter in terms of high pressure zone was first reported by Fyke, Code and Schlegel (1956). Using small pressure transducers, they were able to record the high pressure zone which relaxed in response to a swallow and contracted on the arrival of the

oesophageal peristaltic wave and it was concluded that the high pressure zone is a physiological sphincter.

The high pressure zone is interposed between the stomach and oesophagus and has the characteristics of a valve which offers low resistance to the forward moving but offers a high resistance to retrograde flow in spite of large pressure gradients (Dornhorst et al., 1954).

The sphincteric zone is segmental and at rest lies within or immediately above the diaphragmatic hiatus (Ingelfinger, 1958). It is not certain however that its anatomic position is fixed: the level of maximum sphincteric activity perhaps shifts position under various conditions of oesophageal, gastric and diaphragmatic function. Wankling, Warrian and Lind (1965) reported that the zone of high pressure was equally distributed above and below the diaphragm. This observation was subsequently endorsed by Lind et al. (1966) and Skinner and Camp (1968). The position of canine LOS has also been studied by Rayl, Balison, Thomas and Woodward (1972) by employing combined radiography, manometry and histology. Radiologically, the sphincter is located at a mean distance of 1.87 cm below the abdominal margin of the oesophageal hiatus; manometrically, the sphincter (high pressure zone) extends 0.52 cm above and 0.75 cm below the squamo-columnar epithelial junction, and the two layers of striated muscle are changed into smooth muscle at the level of the sphincter.

In man, the normal resting pressure of LOS measured by open tip catheter varies from 11.0-26.37 mm Hg (Wankling et al., 1965; Waldeck, Jennewein and Siewert, 1973; Csendes, Strauszer and Uribe, 1975; Raizman, Neva, Eckert, Duffy and Lipshutz, 1975). In the dog, this pressure is 11.0 mm Hg (Rosin, Galphin and Bowen, 1979).

The LOS relaxes as the primary peristaltic waves approach it following a deglutition (Code et al., 1958; Ingelfinger, 1958; Pert et al., 1959). This sphincteric relaxation has been found to occur immediately before its contraction (Silber, 1968).

The length of LOS in humans as determined by manometry varies from 0.5-3.4 cm (Dornhorst et al., 1954; Waldeck et al., 1973). In the opossum, this length is 1.3 cm (Lipshutz and Cohen, 1971) and in the dog it is 1.27 cm (Rayl et al., 1972). The length of LOS however, is increased following pentagastrin injection (0.6 $\mu g\ kg^{-1}$ i.v.) from 3.9-5.3 cm (Waldeck et al., 1973).

In sheep, the information is scarce about the length and position of LOS. Manometrically, the high pressure zone has not been demonstrated in this area (Winship et al., 1964). The EMG study of Titchen (1979) however indicated that the lower oesophagus within 1.5-2.0 cm of hiatus represents LOS characteristics but these EMG recordings were unaccompanied by manometric measurements.

Control of LOS

Lower oesophageal sphincter is a unique target organ for many reflex activities. It is also responsive to various gastro-intestinal hormones.

Weisbrodt (1974) suggested three possible mechanisms for LOS closure: tonic activity of nerves innervating sphincter muscle; blood-born substances acting on the sphincteric muscle and activity inherent in the muscle cells themselves. Evidence in support of each of these is presented below.

Roling, Farrell and Castell (1972) find that urecholine increases the LOS tone with a peak response occurring at 35 minutes after subcutaneous injection. The possible mechanism is that this cholinergic agent releases endogenous gastrin which in turn augments the sphincter pressure. Secretin blocks the gastrin-dependent effect of urecholine and the cholinergic response of the LOS is in some part mediated through endogenous gastrin. Similar results have been obtained by Hollis, Levine and Castell (1972). These authors also studied the effect of pentagastrin with doses ranging from 0.1-4.0 mg kg^{-1} subcut and reported that maximum LOS tone is achieved at 4.0 mg kg^{-1} and that the role of gastrin to maintain LOS pressure is physiological. Castell and Harris (1970) also demonstrated in man that endogenous gastrin release resulting from gastric alkalization and sub-maximal subcutaneous dose of pentagastrin (1 μg kg^{-1}) are associated with increased LOS pressure and concluded that stimulation of the LOS appears to be a physiological action of gastrin. It is however reported in the dog by Hollenbeck, Maher, Wickbom, Bushkin, McGuigan and Woodward (1975) that increased serum gastrin resulting from feeding is not associated with any significant change in LOS pressure and suggested that gastrin is not the only hormonal regulator of the LOS pressure. Therefore the majority of the investigators favour the possible role of gastrin in regulating LOS pressure.

It is reported that antigastrin antiserum reduces the LOS pressure by over 70% which suggests that gastrin plays a major physiological role in determining the genesis of basal LOS pressure (Cohen, 1975).

Waldeck et al. (1973) found that LOS pressure after an injection of pentagastrin (0.6 μg kg^{-1} i.v.) changes from 13.0-44.2 mm Hg after

one minute of injection. A significant increase in LOS pressure following intravenous administration of pentagastrin is also reported by Farrell, Castell and McGuigan (1974).

Lipshutz and Cohen (1971) demonstrated in the opossum that the peak response of LOS to gastrin-1 following an intravenous injection, occurs at three minutes and terminates within ten minutes. The maximum response is obtained at a dose of 1.0 $\mu g\ kg^{-1}$. This response of LOS to gastrin is also reported elsewhere in other species (Farrell et al., 1974; Lipshutz, Gaskins, Lukash and Sode, 1974; Cohen, 1975).

The augmentation of LOS pressure in man appears to be independent of the acid secretory stimulus of gastrin (Giles, Mason, Humphries and Clark, 1969). This pressure increase may result from the direct contact of the LOS with the gastric acid which is stimulated by gastrin. Another mode of action of gastrin is reported in the opossum by Lipshutz, Tuch and Cohen (1971) who suggested that gastrin stimulates postganglionic cholinergic nerves to release acetyl choline which in turn increases the LOS pressure. The same mechanism may also exist in man (Lind et al., 1968). On the other hand, Holst, Jensen, Knuhtsen, Nielsen and Rehfeld (1983) found that electrical vagal stimulation increases gastrin output from perfused pig antrum. Kline, McCallum, Curry and Sturdevant (1975) studied in man the effect of alkaline intragastric pH on LOS pressure and on serum gastrin concentration and concluded that intragastric alkalization does not increase total serum gastrin concentration or LOS pressure.

An attempt has been made to correlate the activity of prostaglandins (PG) in the control of LOS. The effects of prostaglandin E1 and E2 in anaesthetized opossums are reported to cause a dose-dependent fall

in LOS pressure (Goyal, Rattan and Hersh, 1973; Dilwari, Newman, Poleo and Misiewicz, 1975). Doses ranging from 0.35-0.4 μg kg^{-1} injected intravenously were found to cause a 50% relaxation in LOS pressure for both PGE1 and E2. The inhibitory effect of PGA2 on LOS pressure is weaker and a 40% drop in LOS pressure is caused by 8.0 μg kg^{-1} in the same route. The hormone perhaps acts as the putative neurotransmitter (ATP) of the inhibitory nerves to the LOS.

In man, it has been found that exogenous application of PGF2α (0.5-0.8 μg kg^{-1} min) produces dose-dependent elevation of LOS pressure while PGE2 inhibits it (Dilwari et al., 1975). The inhibitory effect of PGF2α is not due to the release of gastrin as the circulatory gastrin level remains unaffected by prostaglandins.

The work of Daniel, Crankshaw and Sarna (1979a) in the opossum showed that the development of LOS active tension requires prostaglandin synthesis. Prostaglandin F2α consistently produces increased active tension in LOS and PGE1 and E2 may cause contraction or relaxation. There may be a specialisation of sphincteric muscle to produce a prostaglandin or a precursor (e.g. an endoperoxide) which increases active tension. The responses of sphincteric muscle to E-type prostaglandins may differ from elsewhere in the gut so that they are excitatory when basal tension is low. In a separate work, Daniel, Crankshaw and Sarna (1979b) demonstrated in vitro that muscle strips of the opossum LOS undergo contraction in response to PGF2α which is synthesized by the sphincteric muscle to maintain active tension. Prostaglandin $F_1\alpha$ and PGI_2, on the other hand, caused relaxation to such muscle strips. The muscle strips from the oesophageal body however do not show any response to prostaglandins and this might result from

lack of synthesis in the oesophageal body of contractile prostaglandin or of receptors. They concluded that the active tension in the muscle is myogenic.

Lepsin, Koelz, Weiser, Blum and Siewert (1978) investigated in the dog the role of the duodenum in the regulation of LOS pressure and found that peptones traversing the duodenum raised LOS pressure more than peptone in the stomach. They suggested that this increase may be related to hormones, but the class of hormone is undecided. In the pentobarbitone anaesthetized dog, a gastro-intestinal hormone, motilin has been shown to have a role in the regulation of LOS (Aizawa, Hiwatashi and Hoh, 1978).

The extrinsic nerve supply to the LOS are parasympathetic and sympathetic. The parasympathetic fibres originate in the dorsal motor nucleus of the vagus and reach the LOS through the vagus to synapse with intramural neurons (Goyal and Cobb, 1981). The sympathetic fibres also synapse directly with intramural ganglia.

The precise nervous control of LOS remains undecided. Burget and Zeller (1936) investigated in unanaesthetized dogs the response of oesophageal balloon distension and reported that relaxation of the LOS does not depend on extrinsic nerves as the sphincteric inhibition remains unaffected after bilateral vagotomy. These observations indicate that relaxation of the cardia following an oesophageal peristaltic wave is not dependent upon vagus nerves.

In unanaesthetized dogs, the tonus of the LOS or its power of relaxation remains unaffected by the section of sympathetics (Zeller and Burget, 1937). Bilateral vagotomy in the thorax followed by section of the sympathetics also gave similar results. They felt that relaxation

of LOS needs some sort of reflex to be initiated in the oesophagus which indicates dependence of its activity on intrinsic nerves. They commented that either section of the vagi in the thorax leaves connection along the oesophageal wall with the vagus above the section that is effective in carrying out the reflex, or a myenteric plexus in the oesophagus with which the vagus above the section makes contact and by which it relays impulses to the LOS.

In the opossum, vagal innervation to the oesophagus is responsible for LOS relaxation and unilateral vagotomy does not significantly cause LOS relaxation while bilateral vagotomy eliminates all LOS responses to swallowing (Cohen et al., 1978). Rattan and Goyal (1974) were in agreement with the evidence of Cohen et al. (1978) but recorded an increase of LOS pressure by 113.8% after two minutes following bilateral vagotomy. Electrical excitation of the peripheral end of the left vagus after bilateral vagotomy causes LOS relaxation which lasts longer than central vagal stimulation and later returns to prestimulation base line pressures. On the other hand, electrical stimulation of the central end of the vagus after bilateral cervical vagotomy causes LOS contraction and the pressure increases by 53.1%. This observation suggests that the efferent fibres of the reflex which mediate LOS contraction are not carried in the vagi. They concluded that vagi carry purely inhibitory efferents to the LOS and afferent fibres are involved in a centrally mediated reflex for LOS contraction. However, bilateral stimulation of intact cervical vagi in sheep results in tight closure of LOS (Habel, 1956). Mann and Hardcastle (1968) demonstrated in man that abdominal vagus nerves do not play a major role in controlling the LOS pressure. The parasympathetic branch has an influence in maintaining

the resting LOS pressure since the administration of cholinergic drug significantly increases the resting tone of the sphincter (Heitmann and Espinoza, 1969; Heitmann and Möller, 1970). It is reported that significant elevation of LOS pressure resulting from the administration of metochlopromide is probably mediated by an intramural cholinergic nervous system (Stanciu and Bennet, 1973; Laitinen et al., 1978; Punto, Mokka, Kairaluoma and Larmi, 1977).

Despite the conflicting effects of vagotomy on LOS pressure vagal activity may participate in the maintenance of resting tone since atropine usually reduces the intraluminal pressure (Roman and Gonella, 1981). Different workers however maintain different views about the effect of atropine on LOS pressure. Lind et al. (1968) observed in man that the administration of atropine (0.015 mg kg^{-1}) does not completely obliterate the zone of high pressure and suggested that under resting conditions the LOS pressure is only partially controlled by a cholinergic mechanism operating through the vagus nerves and the remaining pressure in the sphincter may be due to inherent tonicity of the smooth muscle. A fall in resting sphincter pressure resulting from the administration of atropine has been reported by the majority of the workers (Lind et al., 1968; Skinner and Camp, 1968). This fall in sphincter pressure may explain the inhibitory effects of vagus nerve on LOS. Atropine only blocks the vagal excitatory pathway while vagotomy also suppresses the inhibitory one (Roman and Gonella, 1981).

The results of sympathetic nerve section or their pharmacological blockade on LOS are also conflicting. Roman and Gonella (1981) stated that sympathetic denervation does not indicate any clear change in the

sphincter resting tone; but Ingelfinger (1958) reported a slight decrease after denervation. More recent studies in the opposum show that adrenergic denervation by 6-hydroxy dopamine results in decreased basal pressure (Di Marino and Cohen, 1974) and that phentolamine, an adrenergic alpha-receptor antagonist, prevents the transient sphincter hypertension caused by vagotomy (Rattan and Goyal, 1974). On the other hand, sphincter relaxation during swallowing remains unaffected by sympathectomy (Di Marino and Cohen, 1974).

The in vitro study of Waller, Misiewicz, Anthony and Gummer (1971) on isolated muscle strips of human LOS showed that the sphincteric circular muscle contains both adrenergic alpha and beta receptors, the α-receptors being excitatory (cause contraction) and β-receptors inhibitory (cause relaxation). They reported that the inhibitory receptors are mainly present in the longitudinal muscles of the sphincter. The inhibition of resting LOS tone from the stimulation of β-receptors has also been reported by Zfass, Prince, Allen and Farrer (1970) and Di Marino and Cohen (1974).

Epinephrine and nor-epinephrine cause contraction of the LOS in vivo and in vitro (Ingelfinger, 1958; Christensen, 1970; Goyal and Rattan, 1978; Gonella, Niel and Roman, 1979). These motor effects involve α-adrenergic receptors since they are also produced by phenylephrine and blocked by phentolamine (Di Marino and Cohen, 1974).

Investigation in the cat showed that there is a close cholinergic link in the sympathetic control of LOS which is demonstrated by the release of labelled acetyl choline by LOS muscular strips under the action of nor-epinephrine (Gonella, Niel and Roman, 1980).

Electrical excitation of the intact cervical sympathetic nerve in the cat and opossum has no response on the functions of LOS (Fournet, Snape and Cohen, 1978). They also reported that splanchnic stimulation gives an (efferent) increase in LOS pressure and an (afferent) decrease in LOS pressure and that (efferent) excitatory effects on the LOS modulate vagal induced LOS relaxation. The control of LOS tone by adrenergic supply, if there is any, appears to be mainly excitatory (Roman and Gonella, 1981).

Miolan and Roman (1973) suggested that the vagal inhibitory fibres of LOS are preganglionic fibres that probably activate intramural inhibitory neurons, whereas the vagal excitatory fibres are preganglionic fibres which very likely synapse with intramural excitatory neurons (cited by Roman and Gonella, 1981).

In the opossum, the relative sparsity of the myenteric plexus in the area of LOS is consistent with the idea that LOS depends for its physiological character more upon special properties of the muscle than upon a unique or special innervation (Christensen, Rick, Robinson, Stiles and Wix, 1983). The *in vitro* study of Mann *et al.* (1968) demonstrated that the isolated parts of the lower oesophagus along with its sphincter of the guinea-pig and kitten maintained in Tyrode's solution at 36-37°C undergo changes in tone of LOS in response to mechanical distension. This independence from CNS control of LOS in man is also reported by Dornhorst *et al.* (1954) and Pert *et al.* (1959).

Tonic contraction of the LOS is a function of its smooth muscle rather than its control mechanism (Thomas, 1981). He suggested that LOS control mechanism - both nervous and hormonal - act by altering the myogenic responsiveness of the sphincter and such control could be

mediated by varying the level of the resting membrane potential. His illustrations about the nervous innervation and neuro-humoral control of LOS are shown in Figure 2.5.

Fox and Daniel (1978) reported from their *in vitro* study in the opossum that the resting tone of LOS depends on transmembrane Ca^{++} influx resulting from an appreciable inward Ca^{++} leak at rest.

Changes in blood pressure do not seem to have any effect on LOS pressure since the sphincteric pressure has been found to remain unaffected by acute arterial hypotension produced by haemorrhage in anaesthetized opossums (Goyal *et al.*, 1973).

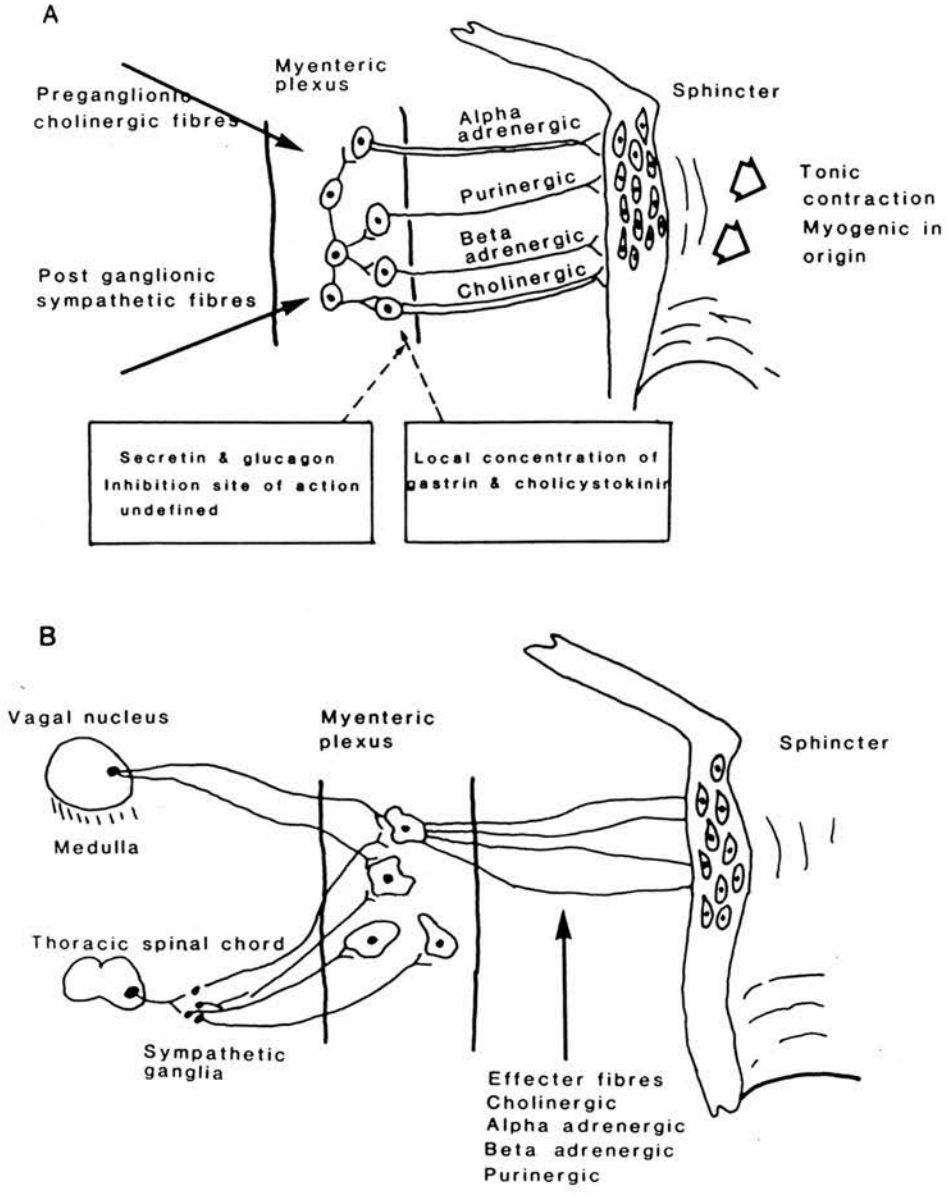
In ruminants, the closure of the LOS is not so much by the tonic contraction of its own musculature, but more by external pressure exerted by surrounding organs on the orifice itself and on the abdominal portion of the oesophagus (Weiss, 1953).

When histological sections of LOS and the body of the oesophagus were studied in the opossum by Christensen and Roberts (1983), they found that the sections of LOS had greater concentrations of mitochondrial mass than the oesophageal body and they postulated that the contractions of LOS muscle is aerobic while those in the oesophageal body may be maintained anaerobically. It is interpreted that the greater mitochondrial mass in the sphincter muscle would be consistent with the greater oxygen dependence of tonic contraction in the sphincter.

Summary

The LOS exists in various species of animals at the region of gastro-oesophageal junction. There has been however very limited amounts of work to investigate this sphincter in ruminants and there is

FIGURE 2.5
Neuro-humoral control of lower oesophageal sphincter (Thomas, 1981).



no concrete evidence of the presence of LOS in this species. Most of the work is on monogastric animals particularly the opossum and the evidence for its occurrence, regulation and control are conflicting.

The nerves supplying the LOS are both intrinsic (myenteric plexus) and extrinsic (vagus and splanchnic). The control mechanism of LOS appears to be complex involving these innervations. The gastrointestinal hormones (e.g. gastrin, prostaglandins) and the sphincteric musculature are also believed to regulate the LOS.

The LOS is regarded as a physiological barrier which prevents the flow of gastric contents up into the oesophagus. In ruminants, however, this sphincter is reflexly relaxed during the regurgitation phase of rumination.

The intraluminal pressure of LOS is greater than that of the stomach and this LOS-gastric pressure gradient is essential to prevent reflux. During anaesthesia, the control mechanism of LOS may be modified or altered and thus predisposes the animal to reflux.

RESPONSES OF LOS TO INCREASED INTRAGASTRIC PRESSURE

The lower oesophageal sphincter is a pressure barrier between stomach and the oesophagus and is involved in preventing the gastro-oesophageal reflux. The pressure maintained in the sphincter is not uniform and is variable depending on the variation of intragastric pressure (Wankling et al., 1965; Diamant and Akin, 1972). They reported that LOS pressure is maintained about 6.0 mm Hg above that in the stomach but the intra-oesophageal pressures remain unchanged.

Gastro-oesophageal reflux occurs when the intragastric pressure rises above that which the LOS can withstand (Marchand, 1955).

Marchand (1955) reported that the reflux of gastric contents into the oesophagus would be continuous if the LOS was incompetent and that this competence in man is substantially contributed by the oblique entrance of the oesophagus into the stomach. He also reported that the diaphragm cannot constrict the terminal oesophagus and that it plays no part in preventing reflux. Pressures ranging up to 11.0 mm Hg can be applied to the abdomen without producing any reflux. Under anaesthesia, intragastric pressures of 7.0-14.0 mm Hg did not overcome the gastro-oesophageal barrier (O'Mullane, 1954).

The increase in tone in the gastro-oesophageal sphincter in response to increased intra-abdominal pressure protects the oesophagus against gastric reflux (Crispin, McIver and Lind, 1967; Diamant and Akin, 1972).

Lind et al. (1966) investigated in man the response of the gastro-oesophageal junction to increased intra-abdominal pressure and showed that increases in intragastric pressure result in a rise in pressure in the junctional zone greater than intragastric pressure indicating the presence of an intrinsic sphincter. The increased pressure in the LOS may be due to inherent tonicity of its smooth muscle (Lind et al., 1968). The majority of workers think that this response of LOS is mediated through a cholinergic reflex arc involving the vagus nerves (Crispin et al., 1967; Lind, Cotton, Blanchard, Crispin and Dimpolo, 1969; Behar and Kastendieck, 1974). A more recent work, however, suggested that it is independent of vagus (Behar, Biancini and Kerstein, 1976). The afferent stimulus for this reflex is presumably the increase in intragastric pressure producing local stretch forces on the smooth muscle of the sphincter and as a result, the LOS muscle contracts to

counteract the stretch forces and thus the pressure is increased (Thomas, 1981). Dodds, Hogun, Miller, Stef, Arndorfer and Lydon (1975), however found no relationship between sphincteric and intragastric pressure.

LOWER OESOPHAGEAL SPHINCTER COMPETENCE AND REFLUX

The mechanisms maintaining the competence of gastro-oesophageal sphincter is controversial (Laitinen et al., 1978). Under those circumstances which favour reflux of gastric contents into the oesophagus, the gastro-oesophageal sphincter responds to prevent or minimise reflux (Diamant and Akin, 1972).

Physiologically, gastro-oesophageal reflux occurs as a reflexly co-ordinated mechanism associated with marked rumeno-oesophageal pressure gradient in sheep (Winship et al., 1964), but pathologically reflux appears to be related with sphincteric strength (Giles et al., 1969). It is reported that the occurrence of reflux is dependent on the ability of the sphincter to maintain competence at the gastro-oesophageal junction (Wankling et al., 1965). However, Skinner and Camp (1968) found in man that there is no correlation between the intraluminal pressure characteristics in the LOS and the incidence of reflux. There are apparently no reports in the literature concerning the competence of LOS, if it exists, and the incidence of reflux of rumen contents in the anaesthetized ruminant.

Fyke et al. (1956) reported in man that the LOS pressure combats gastro-oesophageal reflux. They found that in head-down position which is favourable for reflux by the influence of gravity, the pressure within the LOS becomes more positive to gastric pressure. The tone

of LOS is also increased following contraction of the gastric fundus (Magee, 1962).

The manometrically observed high pressure zone at the gastro-oesophageal junction prevents the occurrence of reflux and the incidence of reflux is always associated with significantly low sphincter pressure (Kelly, 1965; Haddad, 1970). The resting pressure in the LOS is inhibited following a swallow and 90% of reflux is associated with swallow-related LOS relaxation in patients with reflux oesophagitis symptoms (Corazziari, Bontempo, Anzini and Torsoli, 1984). They reported that reflux is more frequently associated with LOS relaxation due to swallowing and less frequently due to lower resting sphincter tone.

Intravenous injection of atropine (0.01 mg kg^{-1}) in anaesthetized dogs reduces the LOS pressure significantly and as a result LOS-gastric pressure gradient almost completely disappears and creates favourable conditions for gastro-oesophageal reflux (Laitinen et al., 1978). The LOS pressure has been considered to correlate directly with sphincter competence (Pope, 1967; Winans and Harris, 1967; Haddad, 1970; Cohen and Harris, 1971). It is suggested that LOS incompetence in patients with gastro-oesophageal reflux may result from diminished gastrin release as well as a defect in the sphincter muscle (Farrel et al., 1974).

RETICULORUMEN MOTILITY AND THEIR CONTROL

Continuous motility of the forestomach is essential in ruminants for a variety of reasons, such as mixing, propulsion, eructation, regurgitation etc. The motility of rumen occurs every 40-60 sec at

rest (Clerk, 1950) while that of the reticulum occurs every 48 sec (Balch, 1961).

There are two sequences of reticulorumen motility: a backward moving peristaltic contraction associated with reticular contraction and a forward moving antiperistaltic eructation contraction not associated with reticular contraction (Kay, 1983). The essential rhythm of reticulum consists of a biphasic contraction (Phillipson, 1939; Duncan, 1951; Leek, 1969; Ehrlein, 1979). These two phases of reticular contraction are of about equal duration (2-3 sec) and the total duration is 4-6 sec (Titchen, 1968).

There are three major factors in controlling reticuloruminal motor activities. These are (1) central and reflex mechanisms operating through gastric centres in the medulla oblongata (Harding and Leek, 1972), (2) intrinsic mechanisms dependent on the myenteric plexus of the rumen wall (Ruckebusch, Tsiamitas and Bueno, 1972; Gregory, 1982), (3) hormonal mechanisms influencing both central and intrinsic control (Grovum, 1981).

The gastric centres associated with the vagal nuclei appear to comprise both rate and amplitude circuits which control impulse discharge by motor neurons leading by the vagus nerves to the reticulorumen (Harding and Leek, 1972). The centres are subject to both excitatory and inhibitory influences, originating both centrally and peripherally. Stimulation of the stomach itself is effective in controlling its motility, moderate stretch and tactile stimuli of the reticulorumen and acidity of the abomasum being excitatory, and severe stretch including distension of the abomasum or acid ruminal conditions that promote rapid volatile fatty acid absorption, being inhibitory.

The *in vitro* experiment of Miert and Huisman (1968) demonstrated that the ruminal smooth muscle in sheep contains α and β-receptors and that the α-receptor is excitatory and the β-receptor is inhibitory.

Environmental cold also stimulates reticulorumen motility (Westra and Christopherson, 1976). A reduction of temperature from 21.2°C-1.3°C causes significant increase of reticulorumen contraction.

In sheep without central nervous connections and maintained by infusing a liquid diet, the reticulorumen contractions are first reduced to irregular twitching, but after a few days, vigorous abnormal contractions resume (Gregory, 1982) suggesting that intrinsic factors (myenteric plexus) are involved in the intact animal and support the early work (Duncan, 1953; Habel, 1956; Ruckebusch *et al.*, 1972).

Reticulorumen contractions occur in response to motor control from the vagus nerve (Clerk, 1950; Titchen, 1958). During feeding these contractions are marked because the sensory receptors projecting in the vagus are excited in the pharynx by food stuff. Reticular contractions in decerebrate preparations of sheep, goats and cattle have been evoked, or when already present increased in force and frequency, or both, by stimulation of the central end of an abomasal or a reticulo-omasal branch of the ventral abdominal vagus nerve (Titchen, 1953). Distension of the rumen was also found to be an effective stimulus and it was concluded that reflex contractions of the reticulum may result from the sensory stimuli from the abomasum, rumen, reticulum itself and probably the omasum.

It has been observed by Comline and Titchen (1951) that in the goat, contractions of the reticulum remains after bilateral vagotomy, splanchnotomy and after destruction of the spinal cord in the thoracic

and lumbar region. Reticular contractions are abolished by the injection of atropine while adrenaline produces a contraction of reticulum in decerebrate sheep. Dougherty (1942) found complete paralysis of the rumen for at least an hour in cattle following a subcutaneous injection of atropine (32.4 mg).

Alexander (1969) postulated that contractions of the reticulorumen depend on the integrity of the vagus nerve and these contractions are elicited by the stimulation of various parts of the medulla. The sympathetic innervation to the rumen has no direct influence on the motility (Habel, 1956). In decerebrate sheep, it is reported that there is a region in the brain caudal to the intercollicular plane which can maintain co-ordinated activity of the reticulorumen (Iggo, 1951). Iggo stated that the receptors of the reticulorumen are tension-sensitive and occur "in series" with the ruminal muscle.

The biphasic contractions of the reticulum in sheep are abolished by vagotomy, atropine, adrenaline and surgical anaesthesia (Duncan, 1951; Comline and Titchen, 1961). In decerebrate sheep, Reid and Titchen (1965) observed that different forms of rumen contraction occur with different frequencies of vagus nerve stimulation. It is suggested that different groups of efferent fibres are involved in the two forms of rumen contraction. It has been found that stretch stimulation of the oesophagus increases the frequency of reticuloruminal contractions (Comline and Titchen, 1957; Sellers and Titchen, 1959; Titchen, 1968). The afferents of such reflex activities are probably present in the vagus. The force and frequency of reticular contractions in decerebrate preparations are increased from omasal canal stimulation.

Borgatti and Matscher (1958) reported in sheep that afferent stimulation of mandibular and maxillary branches of the trigeminal nerve initiate and maintain reticular contractions (cited by Titchen, 1968). These nerves may be stimulated by buccal mucosal stimulation. Stimulation of the reticuloruminal mucosa in the area of the cardia appears to increase the rate of reticuloruminal contractions suggesting that the excitatory receptors for this reflex are located here (Stevens and Sellers, 1959). They also reported that the rate of secondary rumen contraction is markedly increased by stimulation of the ruminal nerves or local pressure on the mucosa of the posterior rumen and is markedly decreased by procainization of ruminal nerves or the topical application of anaesthetic to the dorsal rumen mucosa which indicates that an important number of receptors for a reflex influencing the rate of rumen contraction is located in the posterior sacs of the rumen (Stevens and Sellers, 1959).

Complete unilateral vagal deafferentation in sheep produces brief cessation and then a transient decrease in gastric motility and there is a depression to motor response to filling by food (Falempin and Rousseau, 1979). This brings into the picture the involvement of vago-vagal reflex in maintaining the reticulorumen contraction.

Combined stretching of the reticulum and reticuloruminal fold has been found to be the most effective stimulus for contraction (Comline and Titchen, 1957; Titchen, 1960). Ash (1959) found an increased force and frequency of reticulum contractions in conscious animals with the introduction of acetic acid into the abomasum. These responses are probably due to direct action of the acid on receptors (Titchen, 1968). Mechanical stimulation to the abomasal mucosa also stimulates reticular contractions (Titchen, 1958).

Reticuloruminal contractions are inhibited by the infusion of long chain fatty acids into the duodenum (Titchen, 1968). Nicholson and Omer (1983) also studied the effects of fatty acid infusion into the duodenum in sheep and reported that the reticuloruminal contractions are significantly suppressed by the duodenal infiltration of fatty acids at the rate of 3-5 gm/hr. They suggested that the unsaturated fatty acids stimulate the production of cholicystokinin (CCK) which inhibits the contraction.

Grovum (1981) reported that gastrin and secretin decrease reticuloruminal motility. Intravenous administration of pentagastrin (0.2-1.0 $\mu g\ kg^{-1}$) inhibits reticuloruminal motility (Nicholson, 1982). This inhibition of reticuloruminal motility by gastrin may be brought about through the inhibition of centrally mediated pharmacological receptors (Chapman, Grovum and Newhook, 1979).

The rumen contraction and amplitude are greatly reduced by abomasal administration of 10% copper sulphate (Gutowski, 1982). He also demonstrated that intraruminal administration of 5% hydrochloric acid reduces the contraction rate by 75% and the amplitude by 62%.

The serum mineral concentration also influences the reticuloruminal motility. Huber, Wilson, Stattelman and Goetsch (1981) studied the sensitivity of ruminal contractions to hypocalcaemia in sheep and showed that the contractile strength of the rumen is significantly reduced due to hypocalcaemia. They suggested that this ruminal stasis may arise from the failure of neuromuscular transmission (vagus - rumen).

Summary

Reticuloruminal motility in ruminants is important for regurgitation during rumination, eructation, mixing and propulsion of the

ingesta through the digestive tract. The reticulum exhibits biphasic contractions, while the rumen undergoes two different types of contraction - primary and secondary. The primary ruminal contractions (A-cycle) are associated with reticular contractions and are essential for regurgitation. The secondary ruminal contractions (B-cycle) on the other hand, occur independently from reticular contraction and are usually associated with eructation.

The controlling centre for reticuloruminal motility is located in the brain stem and is involved in the co-ordination of reticuloruminal motor activities through various reflexes mainly mediated through the vagus (vago-vagal reflex). The conditions in the reticulorumen and other parts of the gastrointestinal tract are necessary for initiating such reflex activity.

The intramural innervations (myenteric plexus) and gastrointestinal hormones (e.g. gastrin, secretin, cholicystokinin) are also reported to be involved in the control mechanism.

CHAPTER THREE

OBJECTIVES OF THE STUDY

Every anaesthetist involved in ruminant anaesthesia is well aware of the hazard associated with gastro-oesophageal reflux, but no suggestions about the underlying mechanisms are available for them. The investigations reported here were therefore undertaken to explore certain aspects of the gastro-oesophageal reflux in sheep during anaesthesia.

1. The effect of starvation and various postural positions on the occurrence of reflux. Although, starvation is practised by most of the anaesthetists, its impact as far as the reflux is concerned has not previously been evaluated.

2. Characterization of the pattern of the timing, frequency and volume of rumen contents regurgitated during anaesthesia.

3. Investigation of the basic physiological mechanisms responsible for regurgitation of rumen contents from the mouth during anaesthesia. The regurgitation phenomenon in conscious ruminants is closely associated with reticuloruminal contractions. Also, the passage of rumen contents into the oesophagus and then into the oropharynx may involve active oesophageal antiperistalsis. During anaesthesia, however, whether the rumen contents escape actively or passively has not been investigated.

4. Measurement of the gastro-oesophageal pressure gradient during anaesthesia and its effect on regurgitation. In conscious ruminants regurgitation is associated with a pressure gradient across the LOS. Whether this gradient is also involved in anaesthetized sheep during anaesthesia has not been studied.

5. The effect of the depth of anaesthesia on regurgitation. Since general anaesthetics suppress and modify the various physiological mechanisms, the effect of the depth of anaesthesia on the incidence of reflux was studied. Two planes of anaesthesia, light and deep, were used at random to observe their possible influence on gastro-oesophageal reflux. Different anaesthetic agents including both volatile and injectable ones were investigated.

6. The existence of LOS in sheep. The presence of LOS in ruminants is controversial and therefore the nature of the barrier at the gastro-oesophageal junction is unknown. Experiments were conducted utilising manometric methods to attempt to establish the existence or otherwise of LOS in the anaesthetized sheep.

7. The effect of gastrin on LOS. The factors controlling the activities of LOS are unclear. Such knowledge would be essential from the standpoint of controlling reflux. There has been a substantial amount of work carried out to study these factors in monogastric animals particularly in the opossum. These results, however, are conflicting indicating the complex nature of the control mechanism involving extrinsic and intrinsic innervation, hormonal influence and sphincteric muscle itself. Gastrin, a hormone secreted by the gastric antrum has been found to influence the sphincteric tone in monogastric animals. Hence information is required on whether this hormone also plays a role in maintaining the barrier at the gastro-oesophageal junction in ruminants during anaesthesia.

8. The effect of beta blocker on LOS tone. The LOS in the opossum receives both adrenergic alpha and beta receptors. Stimulation and blocking of these receptors with adrenergic agonists and antagonists modify the sphincteric tone. Investigational information is essential to identify the presence of these receptors in the ruminant LOS which in turn might unveil some of the controlling factors. The effect of propranolol hydrochloride on LOS tone was investigated.

9. The effect of atropine on LOS tone. Most workers believe that atropinization diminishes the sphincter tone and increases the risk of reflux. This possibility has not previously been studied in ruminants under anaesthesia.

10. Investigation in anaesthetized sheep of the electrical activities of the oesophagus and its sphincters with a particular reference to gastro-oesophageal reflux. Electromyography (EMG) is regarded as one of the most sophisticated techniques at the present time to study the physio-pathology of muscular organs in the body. This technique has been mostly limited in ruminants to study gastric and intestinal functions in conscious animals (Ruckebusch, 1970; Grivel and Ruckebusch, 1972; Gregory, 1982). Oesophageal EMG during eructation and regurgitation are reported by Carr et al. (1979) and Titchen (1979).

CHAPTER FOUR

GENERAL MATERIALS AND METHODS

In this section, the basic experimental protocols used in the studies are described. When dealing with the individual experiments, any modifications to the basic techniques will be described in the appropriate section.

General Management of the Experimental Sheep

The experimental animals used throughout this project, unless otherwise stated, were mature Scottish Blackface ewes or Scottish Halfbred ewes. The body weight varied from 35-45 kg. Each sheep was identified with a plastic ear tag.

The sheep were housed in a brick built loose box (14 feet by 14 feet) on straw bedding. The maximum number of sheep penned at any one time was ten. Hay and water were provided ad libitum. This diet was supplemented with a small amount of concentrate feed twice a day. Part of the pen could be divided off to restrain a number of sheep (a maximum of 4) either for restriction of movement or to limit the individual food intake. If absolute food intake restrictions were required, the sheep were moved for a limited period of time to individual pens with concrete floors and no straw bedding.

General Anaesthesia

The experimental programmes were organised so that no individual sheep received more than eight anaesthetics and each sheep was rested for at least two weeks between anaesthetics.

The experimental animals were not usually fasted for anaesthesia. No pharmacological agents were used prior to induction of anaesthesia.

For induction of anaesthesia, the sheep was restrained in right lateral recumbancy on the operating table with a slight head down tilt.

Once the animal was lying relatively quietly, a face mask delivering a mixture of oxygen and nitrous oxide was gently placed over the sheep's nose. The mask was attached to a Magill anaesthetic circuit. The flow rates were set at 4 litres of oxygen and 4 litres of nitrous oxide per minute. Again, once the sheep had accepted the face mask, halothane was introduced gradually until the concentration had reached 4%. This concentration was maintained until the sheep reached surgical anaesthesia. At this level of anaesthesia, the pupil was contracted and remained centrally located. The corneal and palpebral reflexes were abolished and the respiration became deep and regular. Induction of anaesthesia by this technique was remarkably free of excitement or struggling. Following the establishment of surgical anaesthesia, the pharynx and larynx were sprayed with a topical local anaesthetic (Xylocaine Spray, Astra Chemicals Ltd., Watford, Herts., England) to block the pharyngeal and laryngeal reflexes.

Intubation

The head and neck were held in full extension and the jaws were held open by pulling them on either side by an assistant. A laryngoscope (Mackintosh blade with Longworth handle, Penlon Ltd., Abingdon, England) was then introduced with its concave part lying over the base of the tongue and the tip of the blade was positioned just anterior to the epiglottis. The laryngoscope blade was then depressed to illuminate the epiglottis and tracheal intubation was performed using a Magill's cuffed endotracheal tube made of red rubber (Leyland Rubber Company, England). The internal diameter of these endotracheal tubes ranged between 11.0-14.0 mm. The tube was introduced through the trachea until

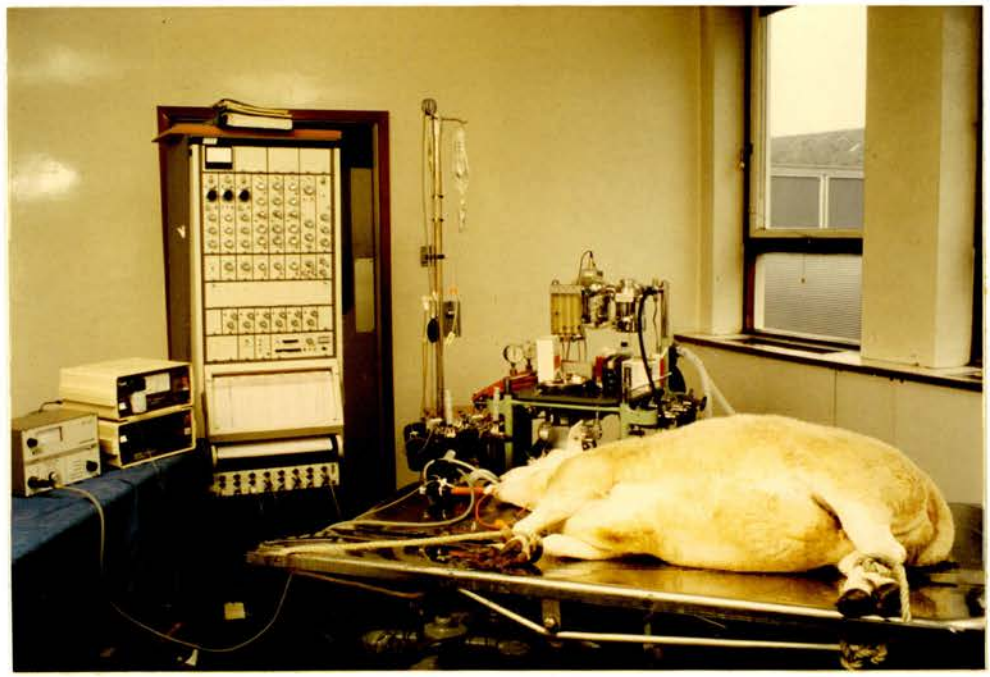
the cuff was behind the larynx. Sometimes the endotracheal tube was introduced using a stilette which was found useful especially in the case of narrow mouthed sheep. The guide (stilette) was withdrawn as soon as the tube was positioned in the trachea. The cuff was then inflated just sufficiently to form an air-tight seal between the tube and the trachea to avoid the risk of aspiration of refluxed rumen material.

Maintenance of Anaesthesia

After intubation, the endotracheal tube was connected to the Boyle's circle carbon dioxide absorber incorporating a sodalime canister. Halothane was introduced to the animal from a "Fluotec-3" calibrated vaporiser (Cyprane Ltd.) through which oxygen and nitrous oxide were delivered at equal flow rates from the anaesthetic apparatus. A steady plane of anaesthesia was maintained by using an expired halothane concentration ranging from 0.75-1.5%. The expired halothane concentration was monitored by an Engstrom Emma anaesthetic gas monitor connected to the expiratory arm of the anaesthetic machine (Figure 4.1). At this level of anaesthesia, there was no palpebral reflex although some degree of corneal reflex was observed. In certain experimental protocols, the influence of depth of anaesthesia was studied. Two planes of anaesthesia, light and deep, were maintained in such experiments. The concentration of expired halothane was maintained between 0.5-1.0% for light anaesthesia which probably represented MAC-1 (Minimum Alveolar Concentration-1). This plane of anaesthesia was characterised by regular breathing and by the presence of palpebral and corneal reflexes. Frequently at this level, visible swallowing could

FIGURE 4.1

Experimental set-up for general anaesthesia. This particular sheep was anaesthetized for measuring the intraluminal pressure changes of the oesophagus and rumen.



be induced by external laryngeal palpation. For the deeper plane of anaesthesia, the concentration of expired halothane was maintained between 1.5-2.0% which probably represented MAC-2. Signs associated with this depth of anaesthesia were deep regular breathing, absence of palpebral and corneal reflexes and absence of swallowing reflex. The pupil was centrally located but still remained contracted. In every case, anaesthesia was continued for at least one hour from the time of induction.

Monitoring of Physiological Parameters

The concentration of oxygen and carbon dioxide in the expired gases and the rate of respiration were measured by an electronic respiratory gas monitor (Datex Normocap^{R} Recorder, Finland). These respiratory parameters were monitored to ensure anaesthetic conditions were maintained as stable as possible.

Recovery from Anaesthesia

On completion of the experimental session, halothane and nitrous oxide were turned off and the oxygen flow was maintained at 4 litres per minute for a period of time. The endotracheal tube was disconnected from the anaesthetic machine. The tube was withdrawn from the trachea as soon as the animal started chewing or jaw movements became apparent. The cuff of the endotracheal tube was not deflated before withdrawal as this might cause aspiration of refluxed material into the respiratory tract and then into the lungs. The animal was then taken to the recovery pen and propped in sternal recumbancy to allow eructation of rumen gases. Once the animal was standing, it was returned to the loose box.

Interval Between the Anaesthetics

Anaesthesia influences the normal physiological and metabolic activities of many of the vital organs of the body, e.g. cardio-vascular system, central nervous system, respiratory system, liver. The extent of these effects may be accumulative if the animals are anaesthetized without adequate time for full recuperation, a reasonable gap of two weeks was allowed between each anaesthetic on an individual animal and it is hoped that this was sufficient for the animal to overcome any adverse effect from the anaesthetics.

The Collection Technique of Refluxed Rumen Material

During anaesthesia, the occurrence, frequency and volume of refluxed rumen material were monitored. The technique employed for collecting the refluxed rumen material involved the use of a second Magill's cuffed endotracheal tube of internal diameter 10.0 mm. After tracheal intubation was achieved, the second tube was introduced into the proximal part of the cervical oesophagus through the oro-pharynx. The tube was introduced until the cuff was posterior to the pharyngo-oesophageal junction. The cuff was then gently inflated (cuff pressure ranging from 100-150 mm Hg). It was hoped that this would prevent the passage of refluxed rumen material between oesophagus and pharynx around the tube to ensure that all refluxed ingesta would flow up the lumen of the collecting tube. Also, the idea of using the cuffed endotracheal tube was to avoid the mixture of refluxed rumen material with the saliva. A polyethylene bag was sealed to the free end of the collecting tube. This bag was supported by a bandage fastened to the polyethylene bag and tied around the horn (if any) or neck so that the

bag was not dragged off the collecting tube when reflux occurred. Where reflux occurred, the volume of the material collected was measured using a laboratory measuring cylinder. The saliva was allowed to run out of the mouth on to the surface of the operating table and then drained into a bucket positioned below the table. The volume of saliva was also measured and recorded. The collection technique is illustrated in Figure 4.2.

Intraluminal Pressure Measurements

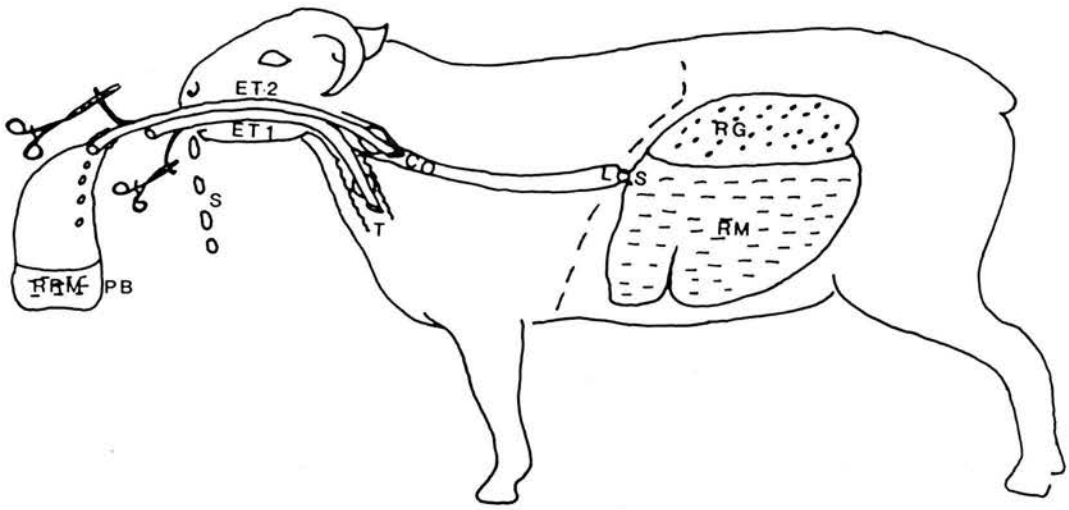
Because the intraluminal pressures of the oesophagus and its sphincter rapidly fluctuate, the equipment required to accurately record the pressure levels must meet more exacting standards than equipment used to measure static pressures. Criteria to be met by apparatus suitable for precise measurement of intraluminal pressures have been discussed comprehensively by Brody and Quigley (1951), Fry (1960), Manktelow and Baird (1969), Stef et al. (1974), Dodds, Stef and Hogun (1976). These criteria will be discussed in some detail at this point as they are also relevant to intraluminal manometry which constitutes a major technique used in this thesis.

There are four major basic requirements to be satisfied for measuring pressure (Manktelow and Baird, 1969). These are sensitivity, linearity, stability and adequate frequency response.

Sensitivity: For the particular pressure being measured, the equipment must give an accurate deflection on the recorder. The range of sensitivity should be approximately 0-750 mm Hg.

Linearity: That the recorded deflection is simply and directly proportional to the pressure load applied to the transducer. This

FIGURE 4.2
Collection technique for refluxed rumen material.



CO. Cervical oesophagus
ET1. Cuffed endotracheal tube (in the trachea)
ET2. Cuffed endotracheal tube (in the cervical oesophagus)
PB. Polyethylene bag
RG. Rumen gas
RM. Rumen material
RRM. Refluxed rumen material
S. Saliva
T. Trachea

linearity is likely to have an approximate error of ± 0.03%.

Stability: (a) With no pressure applied there should be minimal (less than 2% over two hours) base line drift; (b) Repeated application of the same pressure should consistently produce the same deflection.

These three static requirements are usually adequate in modern manometers and amplifiers (Manktelow and Baird, 1969). Stability can be adversely affected however, by allowing inadequate warming up time for the manometer and amplifier or by housing these instruments in an environment with a fluctuating temperature (Fry, 1960; Manktelow and Baird, 1969). Fry (1960) also suggested that the manometer should be minimally handled to prevent finger heat induced changes in the manometer's stability.

Frequency response: The most difficult requirement to obtain is an adequate frequency response (Manktelow and Baird, 1969). The natural frequency of modern strain gauge manometers is very high, up to 1,000 hertz, which is more than adequate for biological pressure measurements, but the necessary fluid filled catheters and connecting tubes greatly reduce the natural frequency of the entire catheter-manometer system. Among the factors that decrease the frequency response of a catheter-manometer system are the presence of air-bubbles within the system, long and narrow catheters and connecting tubes, stopcocks, compressible and viscous fluid within the catheter system, and elastic walls on the catheter system (Manktelow and Baird, 1969).

Manometer Damping

If the membrane of a manometer is displaced by the application and sudden release of pressure, the membrane will vibrate at its own natural frequency (Mendel, 1968). If this same procedure is performed with a fluid filled catheter system attached, the diaphragm and fluid column will now vibrate at the natural frequency of the catheter-manometer system. In practice, however, friction between the fluid and the catheter will cause some damping of the transmitted waves in the fluid and so it is more correctly termed a damped natural frequency (Mendel, 1968).

If the catheter-manometer system records a pressure wave which has a major frequency component similar to its own natural frequency, the entire system will resonate and consequently the recorded pressure wave will be distorted (Grossman, 1974). To prevent resonance, all catheter-manometer systems need extra damping. This damping can be achieved either by (i) a physical constriction along the catheter system or by (ii) stepwise electronic filtering of the manometer output by the pressure amplifier unit (Mendel, 1968).

Theoretically, damping by means of a constriction in the catheter system is the more accurate as it prevents any resonance from developing, whereas electronic filtering allows the catheter-manometer system to resonate but removes the effect of this resonance by damping the recorded signal.

Electronic filtering is more convenient and more widely used because unlike constriction damping, it can be changed stepwise to adapt for different lengths or widths of catheter. A system can, however, be easily overdamped and this will decrease its natural

frequency below optimum (Mendel, 1968). Careful selection of damping scale was used to remove high frequency pen oscillations.

Connecting Fluid

The purpose of the column of fluid within the catheter-manometer system is to transmit by its own movement, pressure fluctuations from the catheter tip to the manometer membrane. Connecting fluid is usually physiological saline (0.9% solution). The most important requirement of the connecting fluid is that it should be non-compressible and in practice this means absence of any air-bubbles (Fry, 1960; Mendel, 1968). This is achieved by boiling the fluid before use to remove its air content and then by slow, continuous flushing under low pressure of the catheter-manometer system to remove any small air bubbles from both the catheter and the manometer (Mendel, 1968).

The use of air as connecting fluid is not able to produce accurate sensitivity, because it can be compressed within the catheter-manometer system. On the other hand, a column of liquid is not compressible but renders inaccuracy with respect to the position of the transducer and the recording tip inside the body. Air in the catheter-manometer system causes damping by lowering the natural frequency of the system (Manktelow and Baird, 1969). In addition, small amounts of air may induce resonance in the system (Mendel, 1968) and if this is similar to naturally occurring events, recording artifacts are produced which are difficult to interpret.

Catheters

The catheters and connecting tubes which transmit the pressure fluctuation from the catheter tip to the manometer membrane must be

non-elastic, otherwise the wall would absorb some of the transmitted pulsations and distort the recording (Fry, 1960; Mendel, 1968).

Hydrostatic Baseline

When measuring pressure by using a liquid filled catheter-manometer system, the recorded pressure is the absolute pressure within the transducer ± the pressure of a saline column equal to the vertical distance between the catheter tip and the mid point of the manometer membrane.

To objectively compare the results of different workers using liquid filled manometers to measure pressure, all must use a common hydrostatic baseline reference point. For this purpose, the transducers and the open tips of the catheters should be at the same horizontal level, so that the pressures detected are due to intraluminal alterations in pressure and not to displacement of the open tip above or below the level of the transducer.

Equipment

Fluid filled pressure transmitting catheters were connected to a strain gauge pressure transducer (type 4-422-0001, SER L74217 and L100888, Bell and Howell Ltd., England). The pressure transducer was connected to a pressure amplifier (3552, Devices Instruments Ltd., Herts.). The results were recorded on a hot stylus multichannel recorder (M19, Devices Instruments Ltd., Herts.). The recorder which was maintained in a laboratory at room temperature was switched on and allowed to warm up for a minimum of two hours before each experiment. The temperature of the connecting fluid was maintained at the temperature of the laboratory to prevent it causing any temperature induced

changes in the catheter-manometer system's damping and stability characteristics (Mendel, 1968). The connecting fluid (tap water) was slowly aspirated into a 20 cc syringe, care being taken to avoid the introduction of any air bubbles. The catheter-manometer system was slowly and repeatedly flushed under low pressure to remove all air bubbles from the system. The system was then calibrated stepwise by electrical calibration signals (full scale deflection = 100 mm Hg). To study the fidelity of the electrical calibration, the system was occasionally calibrated against liquid standards. This involved the use of a column of water in a polyethylene tube connected to a pressure transducer. The other end of the water filled tube was raised to different levels and the deflection was measured.

Catheters

One hundred centrimetre long polyethylene catheters (Portex Ltd., England) of internal diameter of 1.0 mm were used for measuring intraluminal pressures from the oesophagus and rumen. Two types of catheter assembly were employed.

Assembly Type 1: The pressure transmitting catheters were housed in a polyethylene carrier tube of outer diameter 6.0 mm. Three side-holed recording tips were made at 10.0 cm intervals. The distal and proximal ones were open tips and the middle one was a balloon tip. The tip of a condom (Durex, LRC Products Ltd., London) of about 1.0 cm long was used as the balloon which was tied at the tip of the catheter and then glued (Araldite, CIBA-GEIGY, England) to make it air-tight. The tips of the catheters were then glued to the side openings of the carrier tube (Figure 4.3).

FIGURE 4.3

Catheter assembly type 1

a. Open tips

b. Balloon tip (1 .0 cm long) (placed 10.0 cm apart)

c. Polyethylene carrier tube

d. Pressure transmitting catheters

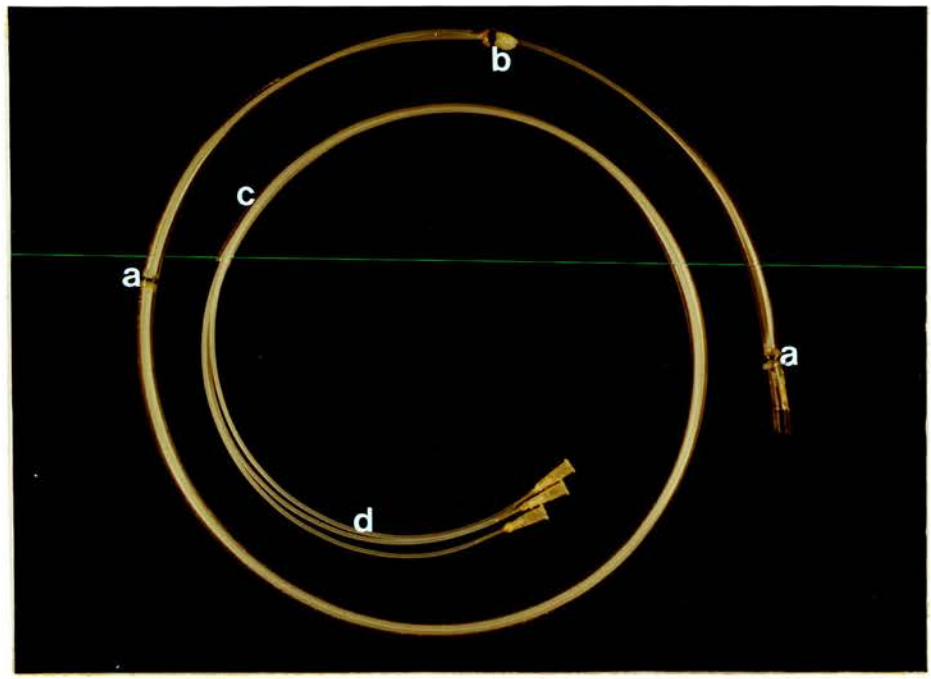
FIGURE 4.4

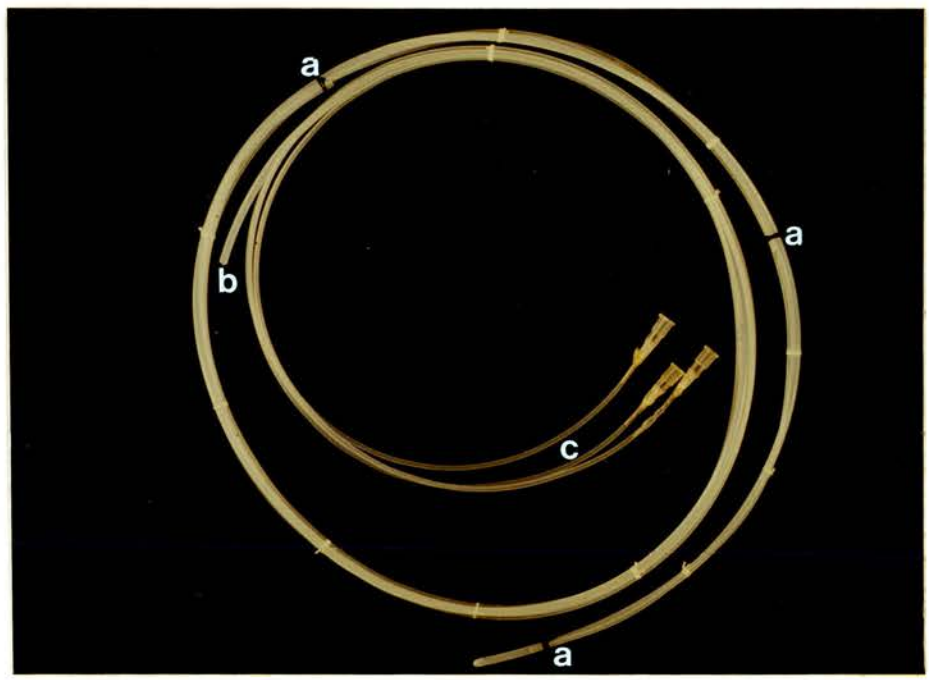
Catheter assembly type 2

a. Open tips (placed 20.0 cm apart)

b. Polyethylene probe

c. Pressure transmitting catheters


b
c
a
a
d


a
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Assembly Type 2: Here the pressure transmitting catheters (all open tips) were assembled together with a solid and flexible polyethylene probe of outer diameter 2.5 mm with the recording orifices situated 10.0 cm apart. The catheters were tied to the probe along its length with cotton thread (Figure 4.4). The free end of the catheter was always connected with 19-gauge needle and glued.

The idea of using the catheter assembly was to position the pressure detecting units at the desired location and also to maintain a uniform distance between the recording tips.

Positioning of the Catheter Assembly

Immediately after induction of anaesthesia, the position of pressure transducers was fixed at the horizontal level of xyphoid cartilage of the animal. The distal end of the catheter assembly was passed through the mouth down to the rumen so that the middle pressure recording tip was also in the reticulorumen. Withdrawal pressure tracings were then recorded as the unit was gradually drawn from the stomach into the oesophagus. The recording system was sufficiently sensitive to register changes of pressure produced by breathing. Within the abdomen, inspiration was marked on the tracing by an upward (positive) deflection while in the oesophagus it was marked by a downward (negative) deflection. The point on the withdrawal tracing at which inspiration changed from a positive to a negative deflection was called "the point of respiratory pressure reversal" and the middle catheter tip was positioned just posterior to that. In this position, the terminal catheter tip was in the rumen approximately 10.0 cm posterior to the LOS, the middle catheter tip in the LOS and the

proximal catheter tip was in the thoracic oesophagus approximately 10.0 cm anterior to LOS. This position was securely maintained by tying the free end of the catheter assembly with the endotracheal tube just anterior to the incisor teeth. After securing the position, the free ends of the catheters were connected with the external electronic pressure transducers using three-way tap. The patency of the catheters was maintained by occasional flushing with water at approximately body temperature. Particular attention was given to remove any air-bubbles present in the catheter-manometer system. In the case of the balloon catheter, 0.2-0.3 cc air was used through the three-way tap using 2.0 ml syringe.

The pressure scales were routinely calibrated before, during and after the experiment to avoid possible drifting from the baseline. Usually, the pressure scales for the rumen and body of the oesophagus were maintained at 25.0 mm Hg and for LOS at 50.0 mm Hg. The pressures from both the rumen and oesophagus including the LOS were always recorded against the atmospheric pressure.

Amplitudes of the oesophageal, LOS and ruminal pressure waves were determined by subtracting mean resting pressures from the peaks of the pressure waves.

The velocity of oesophageal peristalsis was calculated from the time between peristaltic wave peaks recorded in two sites. In this case two open tip catheters were placed 20.0 cm apart along the oesophagus. The duration of a pressure wave was calculated from the time taken to complete the wave i.e. the time taken to reach the peak plus the time to return to the baseline. The speed of the recording paper was usually set at 25.0 mm/min, but for determining the velocity of

oesophageal pressure waves, a higher speed of 25.0 mm/10 sec was used. Other speeds were occasionally used depending on the events to be monitored. The recorded pressure tracings were analysed manually by direct observation.

In the case of the thoracic oesophagus, the amplitude of the respiratory deflections was so large that it was difficult to find a suitable baseline for comparison of pressures at different anatomical levels. This was overcome by choosing the baseline at the midline of the respiratory deflections.

Oesophageal Balloon Distension

The oesophageal response to mechanical distension was studied by distending a balloon in the oesophagus. This study was performed by using a separate catheter assembly incorporated with three polyethylene catheters of internal diameter 1.0 mm and placed 20.0 cm apart. The tip of a condom (Durex, LRC Products Ltd., London) of about 4.0 cm long was used as a balloon, and was tied and glued to the end of the terminal catheter (Figure 4.5). The catheter assembly was introduced through the oro-pharynx into the oesophagus until the balloon lay about 5.0 cm anterior to LOS. This position of the catheter assembly was secured by tying it to the endotracheal tube. The catheter leading to the balloon was left free for the purpose of distension while the other two were connected with pressure transducers to record any changes in the anterior thoracic oesophagus.

The pressure changes of the posterior thoracic oesophagus in response to balloon distension in the cervical oesophagus were also monitored. In this case, the arrangement of the catheter assembly was

FIGURE 4.5

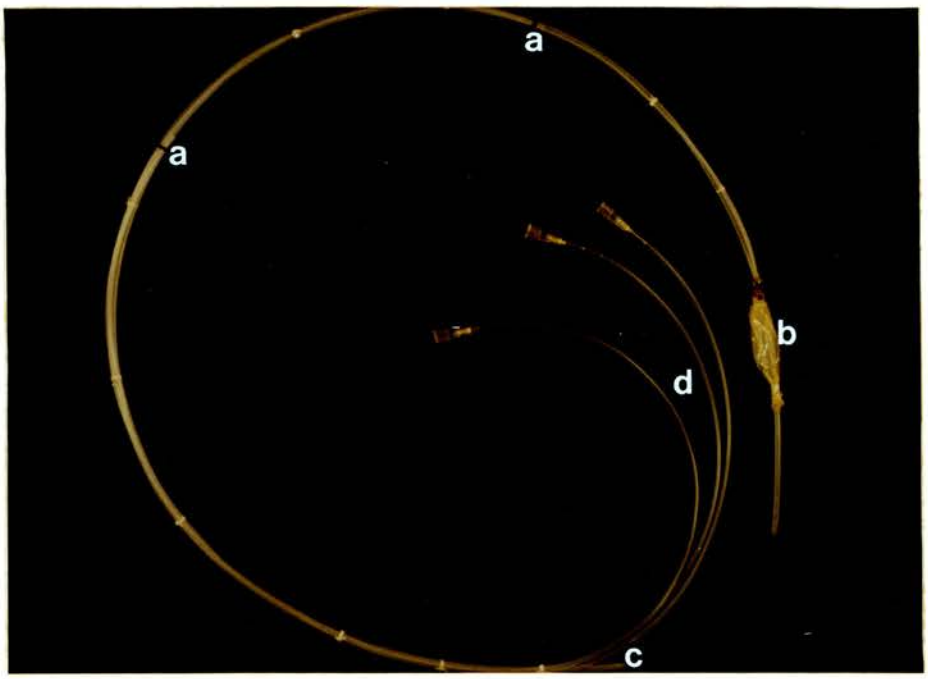
Catheter assembly for intraluminal balloon distension.

a. Open tips

b. Distending balloon (4.0 cm long)

c. Polyethylene probe

d. Pressure transmitting catheters


a
a
b
d
c

modified by changing the location of the balloon from the terminal to the proximal catheter.

After securing the position of the catheter, 25 cc of air was injected into the balloon using a 50 cc plastic syringe. The distension was maintained for 30-60 seconds.

The pressure scales (I) used and shown on the tracings unless otherwise stated represent the relative changes in intraluminal pressure and not the absolute pressure changes.

CHAPTER FIVE

THE INFLUENCE OF DIETARY MANAGEMENT ON GASTRO-OESOPHAGEAL REFLUX

Introduction

It is generally accepted that some form of pre-anaesthetic "starvation" should be practised in ruminant anaesthesia. Anaesthetists use the term "starvation" to mean different degrees of dietary changes e.g. some advocate just withholding concentrates or readily fermentable foods while others advise complete fasting from food and water for a period of time. Hall and Clarke (1983) suggested that the incidence of reflux can be minimised by withholding water for at least six hours before anaesthesia and green foodstuffs and other easily fermentable food for at least 24 hours before anaesthesia.

Pre-anaesthetic starvation in ruminants does not cause complete emptying of the stomach as is the case in the monogastric animals. It minimises the rate of intraruminal gas production and it is suggested that this will reduce the risk of reflux of ruminal contents and minimise the interference to ventilation. These practices are based on clinical impression. There appears to have been no experimental investigation to support these propositions. It was decided to investigate whether starvation has any influence on gastro-oesophageal reflux in anaesthetized sheep.

Materials and Methods

To study the influence of feeding on the hazard of reflux during anaesthesia, 29 sheep were used. The sheep were divided into two groups: an unstarved group and a starved group.

The unstarved group comprised of 17 sheep and included 11 clinical cases. All sheep in this group were housed in a loose box and bedded on straw. These animals were fed hay and water ad libitum. A small

quantity of concentrate ration was fed to the group twice a day. Access to hay and water was permitted up to 10 minutes prior to induction of anaesthesia.

The starved group was made up of 12 sheep. Twenty-four hours before anaesthesia, these sheep were moved from the loose box, where the sheep had been fed in a similar fashion as the unstarved group, to a concrete floored pen. All food and water were withheld for 24 hours.

Sheep from both the groups were anaesthetized in a similar fashion as described in the general materials and methods (Chapter 4). After induction of anaesthesia with a mixture of halothane (4%), nitrous oxide and oxygen (50:50) using a face mask, the sheep were placed in right lateral recumbancy with the operating table adjusted so that the head was tilted down. Anaesthesia was maintained with a mixture of halothane, nitrous oxide and oxygen delivered via an endotracheal tube connected to a Boyle's circle system.

For collecting and quantifying the volume of refluxed rumen contents, a cuffed endotracheal tube was inserted into the proximal part of the cervical oesophagus through the pharynx immediately after induction of anaesthesia. This collecting technique has been described in detail in the general methods (Chapter 4).

Statistical Analysis

Differences in occurrence of reflux between the unstarved and the starved groups were analysed using a Chi-square test and the variation in volume of reflux between these two groups over the 60 minute period of study was calculated by using Mann-Whitney U test.

Results

The experimental observations from the comparison of the two groups (unstarved and starved) are presented in Table 5.1. Depriving the sheep of food and water for 24 hours prior to induction of anaesthesia significantly ($P < 0.05$) reduced the occurrence of reflux. Reflux occurred in 88% of the anaesthetics in the unstarved sheep in contrast to 64% in the starved group. However, although the average total volume* of refluxed materials during anaesthesia was higher for the starved group (572.3 ml/sheep) than in the unstarved group (473.6 ml/sheep), the difference was not statistically significant. The data from each individual experiment is presented in Appendices 5.1 and 5.2.

During the course of this investigation, three patterns of reflux were observed.

(i) Profuse reflux - when large volumes of rumen contents were recorded flowing from the nostrils and mouth over a relatively short period of time.

(ii) Mild reflux - where small volumes of rumen contents were recorded to trickle out of the mouth over a protracted period of time.

(iii) Moderate reflux - medium volumes of gastric contents drained from the mouth and occasionally from the nostrils.

Discussion

An extensive search of literature revealed few experimental studies of the influence of dietary management on the incidence of reflux during anaesthesia in ruminants. In the present study, dietary management has been found to influence the occurrence of reflux during

*These figures on the average total volumes for those anaesthetics where reflux occurred.

Table 5.1 Effect of feeding regime on the occurrence of reflux during halothane anaesthesia (unstarved and starved for 24 hours).

Sheep	Number of anaesthetics	Number of anaesthetics with reflux	Percentage of occurrence	Average volume of reflux (ml)
Unstarved	25	22*	88	473.63
Starved	25	16	64	572.28

*Significant difference ($P < 0.05$)

N.B.

(i) The total volume of refluxed material collected during the 60 minutes of anaesthesia was averaged for those anaesthetics where reflux occurred and this is the figure that is listed under "Average volume of reflux" in the table.

(ii) An animal was considered to produce reflux when the rumen material was drained out from the oro-pharynx.

anaesthesia. Total starvation of the animal of food and water for 24 hours significantly reduced the incidence of reflux. This may be explained as follows. In the starved animal, the rate of intra-ruminal gas production from microbial fermentation may be significantly reduced which may decrease the rumeno-oesophageal pressure gradient across the lower oesophageal sphincter (LOS). This smaller pressure gradient in the starved animal may reduce the potential for reflux of rumen contents through the LOS into the thoracic oesophagus.

It is generally accepted by veterinary anaesthetists that some form of pre-anaesthetic starvation reduces the volume of refluxed rumen material during anaesthesia. However, these studies reveal that the volume of refluxed rumen contents was smaller in the unstarved sheep than the starved sheep although the difference was not significant. There was also a large individual variation in the volume of reflux (5-3580 ml). A possible explanation for this is that the period of starvation makes the rumen contents more fluid (Hecker, Budtz-Olsen and Ostwald, 1964) and as a result when the LOS relaxes, the fluid contents may flow more readily into the oesophagus and then into the oro-pharynx. In the unstarved animal, on the other hand, the relatively solid rumen contents perhaps retard the flow into the oesophagus. The chemical composition of the reticulorumen may also influence reflux: the starved animal having reduced volatile fatty acid levels for example. This possibility, however, was not tested.

Borrie and Mitchell (1960) while discussing pre-operative starving of sheep suggested that reflux is less common after 48 hours of starvation than after 24 hours. This suggests that there is an advantage of longer starvation over the usual period of starvation

(24 hours). A more recent study, however, indicates that prolonged starvation for more than 24 hours has no appreciable effect on reflux (Irwin and Briel, 1966). Irwin and Briel (1966) while studying the dose and effect of pentobarbitone sodium in sheep stated that "Starving for 18-24 hours caused a progressive reduction in both volume and consistency of ruminal ingesta, thereafter the volume diminished slowly if at all, whereas the consistency diminished progressively. Starvation for more than 24 hours increased the hazard of regurgitation without further relieving pressure on the diaphragm or providing more working space in the abdomen". This means that the length of starvation (after 18-24 hours) does not reduce the volume of rumen material as anticipated but makes them more fluid. Thus, the animals after prolonged starvation become more vulnerable to reflux during anaesthesia.

The volume of reflux is relatively less important than its frequency since the danger from reflux during anaesthesia occurs when some of the refluxed material is inhaled. Hecker (1974) reported that the effects of inhalation of rumen contents are not due to a large volume but to an intense bronchospasm. A similar bronchospasm can be produced by instilling 10 ml of water into the trachea of an anaesthetized sheep (Halmagyi and Colebatch, 1962).

Rumen fluid draining from the mouth of the sheep is normally the first sign of reflux seen by the anaesthetist but the reflux from the reticulorumen may occur without any visible drainage. In such cases, the rumen contents move freely along the oesophagus between the two sphincters at either end. In the present study, this was detected with a stethoscope placed over the cervical oesophagus. This provides

evidence supporting the existence of a barrier at the pharyngo-oesophageal junction to the outflow of ingesta.

Three patterns of reflux have been observed in these studies. These are arbitrary groupings, because there were considerable variations between the individual sheep. No attempt was made to record the duration of reflux because of possible difficulties in obtaining accurate measurements. Frequently, the intervals between refluxes were short and it was difficult to clearly define the end point of reflux. Reflux was considered to have occurred when the volume of refluxed material was at least 5 ml. In the present investigation, the amount of reflux was never less than 5 ml. Occasionally, a few flakes of fresh ingesta was noticed within the clear saliva in unstarved sheep. It is likely that these flakes were flushed from the oro-pharynx by the saliva.

From this study, it appears evident that there is a functional barrier at the level of gastro-oesophageal junction. This barrier sometimes becomes incompetent during anaesthesia allowing the entrance of liquid rumen contents into the lower oesophagus driven along the pressure gradient (Hecker, 1974). Incompetence of this barrier may be of varying degrees which may explain the differing patterns of reflux. In the absence of such a barrier, there would be continuous reflux in the head down position.

It is concluded that starvation lasting for 24 hours does not produce any appreciable modification to the mechanism responsible for reflux. Sheep are able to withstand prolonged starvation without any significant change in muscle enzymes or ketone body formation (Mahin,

1983). There are no reports in the literature of the influence of starvation on gastro-intestinal hormones in the sheep.

Conclusions

Gastro-oesophageal reflux during general anaesthesia is regarded as a major hazard in ruminant anaesthesia. This study supports the view that the occurrence of reflux can be reduced by withholding food and water for 24 hours. This result gives support to the general practice that unless emergency surgery is required some form of starvation should be advocated. However, further study is necessary to determine the optimum pattern of starvation or water deprivation which is necessary.

CHAPTER SIX

THE INFLUENCE OF POSITIONING ON GASTRO-OESOPHAGEAL REFLUX

Introduction

Most veterinary anaesthetists consider that positioning may influence the incidence of reflux during ruminant anaesthesia. These views were based on clinical impression and theoretical considerations. There appears to have been no experimental investigations to study the impact of positioning on the occurrence or pattern of reflux during anaesthesia. This study was designed to investigate the problem of reflux during anaesthesia in a number of positions commonly used during veterinary surgery.

Materials and Methods

The effect of positioning on the occurrence of reflux during anaesthesia was studied in 48 sheep. The sheep were divided into the following groups according to position.

(a) Right lateral recumbancy with head down (RLR-HD): This position was studied in 17 sheep. The sheep were placed on the operating table and were restrained in right lateral position for induction of anaesthesia with a face mask. The operating table was adjusted prior to induction so that the head was tilted down at an angle of 15° (Figure 6.1).

(b) Left lateral recumbancy with head down (LLR-HD): The effect of this position was observed in 13 sheep. Induction of anaesthesia was achieved in right lateral position on the operating table with the same degree of tilting (15°) towards the head. Immediately after intubation, the animal was turned to left lateral position (Figure 6.2) which was maintained during the course of anaesthesia.

FIGURE 6.1

Right lateral recumbancy with head down position. The operating table was adjusted so that the animal's head was tilted down (about 15°).

FIGURE 6.2

Left lateral recumbancy with head down position. The operating table was tilted down (about 15°) towards the head.

(c) Dorsal recumbancy with head down (DR-HD): This position was studied in 10 sheep. The animals in this group were first placed in right lateral position on the operating table which was already tilted (15°). Following intubation, the position of the sheep was changed to dorsal recumbancy. This position was achieved by putting two sand bags on either side of the chest while the hind limbs were tied to the operating table with ropes (Figure 6.3).

(d) Right lateral recumbancy with head up (RLR-HU): Twenty-four anaesthetics were performed in this position. Here, anaesthesia was induced in right lateral recumbancy and as soon as the animal was intubated, the operating table was tilted towards the tail of the sheep at an angle of about 20°. In this position, the head of the animal was higher than the xyphoid cartilage. A small sand bag was placed under the neck to ensure that the mouth was lower than the pharynx to allow the drainage of saliva or rumen contents (Figure 6.4).

Sheep were housed in a loose box and bedded on straw and were fed hay and water ad libitum. A small quantity of concentrate ration was supplied twice a day. Feed was available up to 10 minutes prior to the induction of anaesthesia. Sheep from all the groups were anaesthetized in the same manner as described in the general methods (Chapter 4) using a mixture of halothane (1.5-4%), nitrous oxide and oxygen (50:50). In every position, anaesthesia was maintained for a period of 60 minutes.

The refluxed ruminal contents were collected using a cuffed endotracheal tube placed in the cervical oesophagus as described in the general methods (Chapter 4). The data obtained were analysed by using a t-test and a four by two Chi-square test.

FIGURE 6.3

Dorsal recumbancy with head down position. The operating table was tilted down (about 15°) towards the head of the animal.

FIGURE 6.4

Right lateral recumbancy with head up position with the operating table tilted down (about 20°) towards the tail of the sheep.

In a series of control experiments, the influence of the cuffed endotracheal tube in the cervical oesophagus on the occurrence and volume of reflux was studied by comparing the results from five control sheep with no cuffed endotracheal tube in the cervical oesophagus during anaesthesia. Right lateral recumbancy with head down position was maintained during the course of anaesthesia and any occurrence of reflux was recorded over a period of 60 minutes. In this group of sheep, it was not possible to measure the volume of refluxed rumen contents as these were mixed with variable amounts of saliva. The volume of saliva and ruminal contents which occurred was measured at the end of 60 minutes. The data thus obtained were compared with that of RLR-HD position by using a Chi-square test.

Results

The influence of the positioning on the occurrence and average volume of gastro-oesophageal reflux during anaesthesia is summarised in Table 6.1. The dorsal recumbancy with head down position was the most vulnerable situation. In this position, reflux occurred in 100% of cases as compared with right lateral recumbancy with head down position where the occurrence of reflux was 88%.

In left lateral recumbancy with head down position, reflux occurred in 58% of anaesthetics as compared with right lateral recumbancy with head up position where the occurrence of reflux was only 12%.

On statistical analysis, the difference in occurrence of reflux between various postural positions was highly significant ($P < 0.001$).

The volumes of reflux recorded in different positions are presented in Appendices 6.1-6.4. The average volume of reflux in right lateral head down position was 473.60 ml; in left lateral head down position was

Table 6.1 Effect of position on gastro-oesophageal reflux during anaesthesia

(a) Observed values

Position	Number of anaesthetics	Number of anaesthetics with reflux	Percentage of occurrence	Average volume of reflux (ml)
Right lateral head down (RLR-HD)	25	22	88	473.60
Left lateral head down (LLR-HD)	14	8	58	177.90
Dorsal head down (DR-HD)	12	12	100	527.50
Right lateral head up (RLR-HU)	24	3	12	283.30

(b) Significance of differences in occurrence of reflux attributable to position

Positions compared	Degrees of freedom	Chi-square	P	Assessment
All positions	3	89.000	< 0.001	Highly significant
RLR-HD + DR-HD vs LLR-HD + RLR-HU	1	30.948	< 0.001	Highly significant
RLR-HD vs RLR-HU	1	26.411	< 0.001	Highly significant

N.B. All other positions were not valid

(c) The influence of positioning on the average volume of reflux

Positions compared	Kruskal-Wallis H	P	Assessment
All positions	32.595	<0.01	Highly significant differences
All head down positions	9.787	<0.05	Significant differences
Lateral positions	1.945	>0.05	Not significantly different

i.e. The head up position produces significantly less reflux than all the other positions.
Dorsal recumbancy with head down produces significantly more reflux material than the lateral positions.
There is no significant difference in the volume of reflux on either of the lateral positions with the head tilted down.

177.90 ml; in dorsal head down position was 527.50 ml and in right lateral head up position was 283.30 ml. Positioning significantly affected the average volume of reflux (Table 6.1c).

The effect of the presence of cuffed endotracheal tube in the cervical oesophagus on the occurrence of reflux is presented in Table 6.2. In the presence of a cuffed endotracheal tube in the cervical oesophagus reflux occurred in 88% of the anaesthetics in contrast with 54.5% cases in the sheep without a tube and this difference was significant at 5% level. The detailed records are presented in Appendix 6.5.

The insertion of a laryngoscope for intubating the trachea provoked reflux in 32% of cases.

Discussion

On theoretical grounds, gastro-oesophageal reflux during anaesthesia may occur in several circumstances. Firstly, the rumen contents may be carried to the pharynx by active oesophageal antiperistalsis. Secondly, the refluxed material may be driven out by gastro-oesophageal pressure gradient. Thirdly, the rumen contents may be drained out along hydrostatic pressure gradients. The patency of LOS, however, is a common feature in all the above possibilities.

The reticulorumen must obey hydrostatic laws as it is a fluid container. The pressure in these organs in any anatomical point, therefore, depends on the position of the animal. The manometric observation of Botha (1959) with monogastric animals including the dog and cat indicates that "the intragastric pressure is high when the head is down and drops in relation to the resting horizontal level when the head is

Table 6.2 Influence of cuffed endotracheal tube in the cervical oesophagus on the occurrence of reflux during halothane anaesthesia.

Sheep	Number of anaesthetics	Number of anaesthetics with reflux	Percentage of occurrence	Average volume of reflux where occurred (ml)
With tube	25	22*	88	473.60
Without tube	11	6	54.5	-

*Significant difference ($P < 0.05$)

raised. Whereas it is always positive in the supine position, it may change to an absolute negative value when the head is elevated".

In the present study, the occurrence of reflux during anaesthesia was influenced by various positionings. The highest incidence was encountered in DR-HD position. The reasons are not clear but a greater gradient due to hydrostatic pressure is likely to play some role in its occurrence. Also, in this position, the neck of the animal remains relatively stretched which may pull the LOS a little forward. This displacement of the anatomical location of LOS may affect its strength and this may provoke a reflux.

The lowest incidence of reflux was obtained in RLR-HU position. In this position, the pharynx remains higher than the LOS and as a result the difference of pressure between reticulorumen and oesophagus may be partially neutralized by the opposite hydrostatic pressure gradient. Also, the rumen contents if already trickled into the caudal oesophagus are unlikely to flow up along the oesophagus against this gradient in the absence of active antiperistalsis.

The occurrence of reflux was also higher in RLR-HD position than LLR-HD. The reasons are not understood as the intraruminal pressures in these two positions are almost similar.

The insertion of a laryngoscope for intubating the trachea appeared to provoke reflux. The use of a laryngoscope during intubation may mechanically interfere with the closing mechanism of pharyngo-oesophageal sphincter and as a result any rumen contents present in the cervical oesophagus are drained out through the oro-pharynx. Alternatively, this manoeuvre might induce oesophageal antiperistalsis which carries the refluxed rumen contents up to the pharynx. A further

possibility is that a reflex may exist whereby excitation of pharyngeal sensory receptors may cause the LOS sphincter to relax.

The collection technique involved the use of a cuffed endotracheal tube into the cervical oesophagus. To investigate whether the presence of such a tube into the oesophagus provokes reflux, a series of control experiments were performed without a cuffed endotracheal tube in the cervical oesophagus. The incidence of reflux in this group was significantly lower. The increased occurrence of reflux in the group with a cuffed endotracheal tube is probably due to the lack of any resistance at the pharyngo-oesophageal sphincter which cannot retard the flow of ingesta. Therefore, any rumen material that escapes the barrier at the gastro-oesophageal junction has no further barrier to overcome before entering the oro-pharynx. This suggests that the pharyngo-oesophageal sphincter may act as a second oesophageal barrier to the flow of reflux. The presence of a cuffed endotracheal tube in the cervical oesophagus may also provide sensory stimulation for a reflex relaxation of the LOS through vagal efferents which in turn may produce reflux.

It is worth noting that on no occasion where a cuffed endotracheal tube was placed in the cervical oesophagus did ingesta pass on the outside of the cuff of the endotracheal tube. All the ingesta drain down the lumen of the cervical tube. Thus there was virtually no possibility of ingesta entering the pharynx or into the trachea. The presence of a cuffed endotracheal tube in the oesophagus increases the likelihood of reflux, but as it ensures that no refluxed material is carried to the pharynx, this tube can be used with minimum risk.

Evidence during these series of experiments suggests that the reflux of rumen contents during anaesthesia is a passive phenomenon.

Conclusions

Positioning of the sheep during general anaesthesia influenced the occurrance of gastro-oesophageal reflux. This hazard may be minimised by proper management as follows:-

(i) During intubation, the head of the animal should be elevated, because in this position reflux is not likely to occur.

(ii) The animal's position during anaesthesia should be maintained in left lateral recumbancy where possible.

(iii) A cuffed endotracheal tube should be placed in the upper cervical oesophagus to prevent aspiration of rumen contents as it ensures that any ingesta that is refluxed is drained out of the mouth.

(iv) The hind quarter of the animal should be tilted down at an angle of 20° and a sand bag placed under the neck to ensure that the mouth is lower than the pharynx to allow the drainage of saliva or any rumen contents which may be refluxed.

CHAPTER SEVEN

THE INFLUENCE OF GASTRO-OESOPHAGEAL PRESSURE GRADIENT ON REFLUX

Introduction

The mechanism of gastro-oesophageal reflux during anaesthesia is not fully appreciated. It has been widely postulated that among the factors that may be involved are the build up of intraruminal pressure and the pressure gradient across the lower oesophageal sphincter (LOS). An investigation was undertaken of the importance of these two factors on gastro-oesophageal reflux in the anaesthetized sheep.

Materials and Methods

Thirteen sheep were anaesthetized on 33 occasions to study the relationship of the intraruminal pressure gradient between reticulo-rumen and thoracic oesophagus and the occurrence of reflux. Four of these sheep were not starved prior to anaesthesia (24 anaesthetics), and nine sheep were starved (9 anaesthetics).

Induction of anaesthesia was achieved with a mixture of halothane (4%) and oxygen and nitrous oxide (50:50) using a face mask and a Magill system. After induction, the anaesthesia was maintained as described in the general methods (Chapter 4).

The animals used in this study were both unstarved and starved (24 hours) and were positioned in right lateral recumbancy. Pressures in the oesophagus and rumen were measured for a period of 60 minutes by using water filled open tip catheters and employing the catheter assembly 1 as described in the general methods (Chapter 4). The catheter assembly was introduced through the oro-pharynx and positioned along the alimentary canal such that the recording tip for the rumen lay about 10 cm posterior to the LOS and that for the oesophagus about 10 cm anterior to LOS.

Two planes of anaesthesia, light and deep, were maintained for two 30 minute periods in a single anaesthetic session. The selection of the plane of anaesthesia for the first 30 minutes was random. During the light plane of anaesthesia, the concentration of halothane in the expired gases was maintained between 0.5-1.0% by using an Engstrom Emma anaesthetic gas monitor. Clinically, this plane of anaesthesia was characterised by the presence of palpebral and corneal reflex. In the deep plane of anaesthesia, a concentration of halothane ranging from 1.5-2.0% in the expired gases was maintained. The palpebral and corneal reflex were usually absent in the deep plane of anaesthesia.

The influence of these two planes of anaesthesia on the change of intraruminal pressure and the incidence of reflux was studied.

Results

The initial rumen pressure varied considerably between different animals and in individual animals on different days. In all animals, the intraruminal pressure was always positive with respect to atmosphere.

Light followed by deep anaesthesia (60 minutes): The intraruminal pressures recorded in light followed by deep anaesthesia are presented in Table 7.1. The individual records of intraruminal pressure are shown in Appendix 7.1. The intraruminal pressure increased to 9.33 ± 1.51 (S.D.) mm Hg from an initial pressure of 6.78 ± 1.31 (S.D.) mm Hg (an increase of 2.55 mm Hg).

Deep followed by light anaesthesia (60 minutes): The intraruminal pressure records in deep followed by light anaesthesia are

Table 7.1 Intraruminal pressure built up during halothane anaesthesia (light followed by deep)

Sheep No.	No. of trials	Initial pressure (mmHg)±S.D.	Pressure after 10 mins. (mmHg)±S.D.	Pressure after 20 mins. (mmHg)±S.D.	Pressure after 30 mins. (mmHg)±S.D.	Pressure after 40 mins. (mmHg)±S.D.	Pressure after 50 mins. (mmHg)S.D.	Pressure after 60 mins. (mmHg)±S.D.
192D	3	6.16±1.60	7.0±1.80	7.16±2.25	8.0±2.29	8.50±3.50	8.33±2.30	9.50±3.90
243D	3	5.66±1.75	6.0±2.29	6.66±2.30	6.83±2.02	6.83±1.75	8.50±1.80	9.50±1.0
27x91	3	8.66±5.13	10.03±6.10	10.16±5.79	10.83±6.37	11.43±6.0	11.13±6.21	11.0±6.38
289D	3	6.66±4.04	6.66±4.04	7.33±4.04	7.50±4.09	7.0±3.77	7.66±4.16	7.33±4.04
Total	12	6.78±1.31	7.42±1.78	7.82±1.58	8.29±1.75	8.44±2.12	8.90±1.52	9.33±1.51

presented in Table 7.2. The intraruminal pressure increased to 12.62 ± 1.63 (S.D.) mm Hg from an initial pressure of 7.62 ± 1.57 (S.D.) mm Hg (an increase of 5.0 mm Hg). The individual values are shown in Appendix 7.2.

On statistical analysis, the intraruminal pressure built up in deep followed by light anaesthesia was significantly ($P < 0.05$) greater than that in light followed by deep anaesthesia (Figure 7.1).

The intraruminal pressure built up in the first 30 minutes and second 30 minutes in light followed by deep anaesthesia was calculated (Table 7.3). The intraruminal pressure built up in the first 30 minutes was 1.50 ± 0.60 (S.D.) mm Hg and that in the second 30 minutes was 1.04 ± 1.30 (S.D.) mm Hg. The difference was not statistically significant.

The intraruminal pressure built up in the first 30 minutes and second 30 minutes in deep followed by light anaesthesia was also recorded and the results are presented in Table 7.4. The intraruminal pressure built up in the first 30 minutes was 3.49 ± 1.73 (S.D.) mm Hg and that in the second 30 minutes was 1.99 ± 1.47 (S.D.) mm Hg. There was again no significant difference between the two periods.

The pressure gradients between rumen and thoracic oesophagus associated with reflux are shown in Table 7.5. The intraruminal pressure was 8.91 ± 2.97 (S.D.) mm Hg and the intra-oesophageal pressure was 1.73 ± 0.24 (S.D.) mm Hg. The rumeno-oesophageal pressure gradient was 7.18 ± 3.16 (S.D.) mm Hg.

The maximum pressure gradient between rumen and thoracic oesophagus associated with reflux was recorded (Table 7.6). The maximum intraluminal pressures of rumen and thoracic oesophagus were 9.23 ± 3.20

Table 7.2 Intraruminal pressure built up during halothane anaesthesia (deep followed by light)

Sheep No.	No. of trials	Initial pressure (mmHg) ±S.D.	Pressure after 10 mins. (mmHg) ±S.D.	Pressure after 20 mins. (mmHg) ±S.D.	Pressure after 30 mins. (mmHg) ±S.D.	Pressure after 40 mins. (mmHg) ±S.D.	Pressure after 50 mins. (mmHg) ±S.D.	Pressure after 60 mins. (mmHg) ±S.D.
192D	3	6.50±0.86	8.83±0.76	9.93±1.25	12.33±3.05	13.16±3.40	13.50±2.17	12.83±1.04
243D	3	5.66±2.75	6.16±3.21	8.23±2.85	9.16±2.25	9.66±1.60	10.50±1.80	11.66±1.44
27x91	3	9.33±4.04	9.83±4.53	10.0 ±4.27	11.0 ±5.56	12.33±6.50	13.16±7.52	14.83±8.25
289D	3	7.0 ±1.0	7.83±1.60	9.33±1.75	10.0±1.50	10.16±2.75	11.5 ±3.27	11.16±4.01
Total	12	7.62±1.57	8.16±1.56	9.37±0.81	10.61±1.36	11.32±1.68	12.16±1.41	12.62±1.63

FIGURE 7.1

Relationship of intraruminal pressure built up during halothane anaesthesia between light followed by deep and deep followed by light manoever.

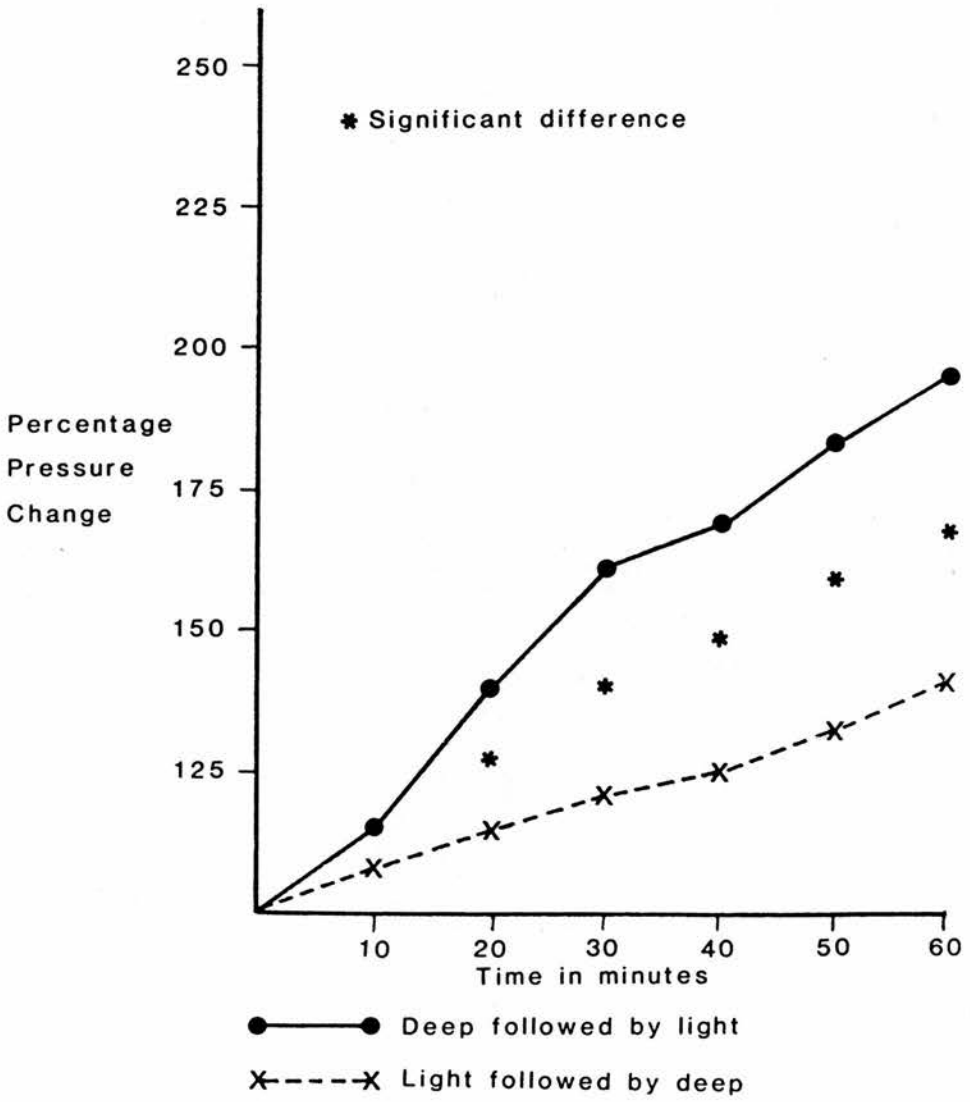


Table 7.3 Relationship of intraruminal pressure built up during halothane anaesthesia (light followed by deep) between the first 30 minutes and second 30 minutes

Sheep No.	Number of anaesthetics	Pressure built up in the first 30 minutes (mmHg)	Pressure built up in the second 30 minutes (mmHg)	Statistical difference
192D	3	1.84	1.50	
243D	3	1.17	2.67	
27x91	3	2.17	0.17	
289D	3	0.84	-0.17	
Mean		1.50	1.04	P > 0.05
S.D.		0.60	1.30	

Table 7.4 Relationship of intraruminal pressure built up during halothane anaesthesia (deep followed by light) between the first 30 minutes and second 30 minutes

Sheep No.	Number of anaesthetics	Pressure built up in the first 30 minutes (mmHg)	Pressure built up in the second 30 minutes (mmHg)	Statistical difference
192D	3	5.83	0.50	
243D	3	3.50	2.50	
27x91	3	1.66	3.83	
289D	3	3.0	1.16	
Mean		3.49	1.99	P > 0.05
S.D.		1.73	1.47	

Table 7.5 Pressure gradient between rumen and thoracic oesophagus associated with reflux during all planes of halothane anaesthesia

Sheep No.	Pressure gradient associated with reflux (mmHg)		
	Rumen	Thoracic oesophagus	Gradient
192D	5.5	5.5	0.0
	5.0	5.0	0.0
	7.0	4.5	2.5
	15.5	3.5	12.0
	13.5	2.0	11.5
	12.0	1.5	10.5
	12.0	2.0	10.0
	12.0	-1.5	13.5
	14.0	2.0	12.0
	7.0	0.0	7.0
	7.0	0.0	7.0
	7.0	0.0	7.0
	7.5	-0.5	8.0
	10.0	2.5	7.5
	12.0	1.0	11.0
	12.5	1.5	11.0
	12.5	3.0	9.5
Mean ± S.D.	10.11±3.30	1.88±1.98	8.23±4.07
243D	10.5	0.0	10.5
	12.0	3.0	9.0
	12.0	2.5	9.5
	12.0	1.0	11.0
	12.5	2.5	10.0
	5.5	0.5	5.0
	7.5	1.0	6.5
	8.5	1.5	7.5
Mean ± S.D.	10.06±2.59	1.5±1.06	8.62±2.10
27x91	4.5	2.0	2.5
289D	8.0	2.5	5.5
	7.5	1.5	6.0
	8.0	-2.0	10.0
	13.0	3.0	10.0
	13.5	3.0	10.5
	15.0	3.5	11.5
	9.5	0.5	9.0
	11.0	1.0	10.0
	11.5	1.5	10.0
	12.0	1.0	11.0
	12.0	1.5	10.5
Mean ± S.D.	11.0±2.46	1.54±1.52	9.45±1.94
Total Mean	8.91	1.73	7.18
S.D.	2.97	0.24	3.16

Table 7.6 Maximum pressure gradient between rumen and thoracic oesophagus associated with reflux during all planes of halothane anaesthesia

Sheep No.	Maximum pressure gradient associated with reflux (mmHg)		
	Rumen	Thoracic oesophagus	Gradient
192D	7.0	4.5	2.5
	15.5	3.5	12.5
	12.0	-1.5	13.5
	7.0	0.0	7.0
	12.0	1.0	11.0
243D	12.0	1.0	11.0
	8.5	1.5	7.0
27x91	4.5	2.0	2.5
289D	8.0	-2.0	10.0
	15.0	3.5	11.5
	11.0	1.0	10.0
	12.0	1.0	11.0
Mean	9.23	1.40	7.83
S.D.	3.20	0.47	3.62

(S.D.) mm Hg and 1.40 ± 0.47 (S.D.) mm Hg respectively. The maximum gastro-oesophageal pressure gradient associated with reflux was 7.83 ± 3.62 (S.D.) mm Hg.

The maximum pressure gradient not associated with reflux was also recorded (Table 7.7). The maximum intraruminal pressure was 8.70 ± 1.47 (S.D.) mm Hg and that of intra-oesophageal pressure was 1.62 ± 0.21 (S.D.) mm Hg. The maximum pressure gradient between rumen and thoracic oesophagus was 7.08 ± 1.32 (S.D.) mm Hg.

On statistical analysis, there was no significant difference in maximum gastro-oesophageal pressure gradient between the anaesthetics which were associated with reflux and those not associated with reflux.

When the number of refluxes in animals anaesthetized by light followed by deep was compared with deep followed by light, there was no significant difference in the incidence of reflux (Table 7.8). However, there were significant differences between sheep in the incidence of refluxes (Chi-square = 13.919, $P < 0.01$). This significant difference was attributable to sheep 27x91 (Chi-square = 3.167, $P > 0.20$).

The intraruminal pressure build up in the starved sheep was significantly lower ($P < 0.01$) when compared with the unstarved group (deep followed by light anaesthesia) but not significantly different from the light followed by deep unstarved group (Table 7.10).

In the unstarved animals, strong rumen contractions were present in light anaesthesia (Figure 7.3A). In the starved animals, however, the rumen contractions were weaker (Figure 7.3B). These contractions were absent in both unstarved and starved sheep in deep anaesthesia.

Table 7.7 Maximum pressure gradient between rumen and thoracic oesophagus not associated with reflux during all planes of halothane anaesthesia

Sheep No.	Maximum pressure gradient not associated with reflux (mmHg)		
	Rumen	Thoracic oesophagus	Gradient
192D	12.5	3.5	9.0
	7.0	0.0	7.0
243D	8.0	3.0	5.0
	8.5	1.0	7.5
	8.5	2.5	6.0
	8.0	2.5	5.5
	10.0	0.0	10.0
27x91	8.5	1.0	7.5
	11.5	5.5	6.0
	10.0	1.5	8.5
	9.0	0.0	9.0
	10.0	0.0	10.0
289D	9.0	1.0	8.0
	8.0	2.5	5.5
	3.0	0.5	2.5
Mean	8.70	1.62	7.08
S.D.	1.47	0.21	1.32

Table 7.8 Total refluxes in sheep anaesthetized three times by light followed by deep and three times by deep followed by light

Sheep No.	Light-deep	Deep-light	Difference
192D	7	10	-3
243D	5	4	+1
27x91	0	1	-1
289D	5	5	0

$t_{[3]} = 0.878$, $P > 0.40$

Table 7.9 Intraruminal pressure built up in sheep starved for 24 hours during halothane anaesthesia over a period of 60 minutes

Serial No.	Initial pressure (mmHg)	Pressure after 10 mins (mmHg)	Pressure after 20 mins (mmHg)	Pressure after 30 mins (mmHg)	Pressure after 40 mins (mmHg)	Pressure after 50 mins (mmHg)	Pressure after 60 mins (mmHg)
1	6.5	7.0	7.0	7.0	7.0	7.0	7.0
2	6.5	7.0	7.0	7.0	7.0	7.0	7.5
3	5.0	5.5	5.5	5.5	5.5	5.5	5.5
4	6.0	6.0	6.0	6.0	6.0	6.5	6.5
5	9.5	9.5	9.5	9.5	9.5	9.5	9.5
6	7.5	7.5	7.5	8.0	7.5	7.5	7.5
7	6.0	6.5	6.5	6.5	6.5	6.5	6.5
8	7.0	7.0	7.0	7.0	7.0	7.0	7.0
9	5.5	5.5	5.5	5.5	5.0	5.0	5.0
Mean	6.61	6.83	6.83	6.88	6.77	6.83	6.88
S.D.	1.31	1.22	1.22	1.26	1.30	1.27	1.29

Table 7.10

(a) Intraruminal pressure built up in the starved group compared with the unstarved group (light anaesthesia)

Time	Starved N = 9	Unstarved N = 4	t	Assessment
0	6.61 ± 1.31	6.78 ± 1.31	0.216	Not significant
10	6.83 ± 1.22	7.42 ± 1.78	0.704	Not significant
20	6.83 ± 1.22	7.82 ± 1.58	1.241	Not significant
30	6.88 ± 1.26	8.29 ± 1.75	1.664	Not significant

(b) Intraruminal pressure built up in the starved group compared with the unstarved group (deep anaesthesia)

Time	Starved N = 9	Unstarved N = 4	t	Assessment
0	6.61 ± 1.31	7.62 ± 1.54	1.213	Not significant
10	6.83 ± 1.22	8.15 ± 1.56	1.675	Not significant
20	6.83 ± 1.22	9.37 ± 0.81	3.764	< 0.01
30	6.88 ± 1.26	10.61 ± 1.36	4.819	< 0.01

FIGURE 7.3

Ruminal pressure changes during light anaesthesia using water filled open tip catheters.

A. Dorsal sac of rumen (10 cm posterior to LOS) in the unstarved animal.

B. Dorsal sac of rumen (10 cm posterior to LOS) in the starved animal.

In the unstarved animal (A), the rumen usually showed strong pressure waves while in the starved animal (B), these pressure waves were usually weaker. A pressure wave in the tracing is indicated by a transient upward fluctuation of baseline pressure and this represents an active contraction.

Discussion

The intraruminal pressure built up in deep followed by light anaesthesia was significantly higher than that during light followed by deep anaesthesia. The mechanism responsible for this difference is not clearly understood. However, the following explanation may be one of the possibilities. Halothane is known to produce hypotension in deep anaesthesia (Wood-Smith, Stewart and Vickers, 1968) and as a result the blood circulation to the walls of the reticulorumen may be depressed and consequently less ruminal gas may be absorbed. This means an increased rate of intraruminal pressure build up. So, in the case of deep followed by light anaesthesia, the intraruminal pressure built up in the first 30 minutes may be faster. In the second 30 minutes, when the light plane of anaesthesia is maintained, the previous trend of intraruminal pressure build up probably continues for a period of time. On the contrary, in the case of light followed by deep anaesthesia, the absorption of reticuloruminal gases is faster in the first 30 minutes because of the relatively unimpaired blood supply to the reticulorumen and this means a slower rate of intraruminal pressure build up. After 30 minutes, when the plane of anaesthesia is changed to deep, the slow fashion of intraruminal pressure build up may still continue for some time. This hypothetical mechanism may help to explain the reason for an increased intraruminal pressure build up in deep followed by light anaesthesia than in light followed by deep anaesthesia.

In the unstarved animal, the intraruminal gas production was relatively faster than that in the starved animal and this could make the gastro-oesophageal pressure gradient greater. This gradient is further

increased by relatively strong rumen contractions in light anaesthesia.

In the starved animals, the rumen contractions are weaker than those in the unstarved animals. The presence of rumen contractions in a light plane of anaesthesia periodically increases the gastro-oesophageal pressure gradient. This gradient in association with slight relaxation of LOS may cause the occurrence of reflux (Bell, 1961).

The significant suppression of intraruminal gas production in a starved animal obviously reduces the gastro-oesophageal pressure gradient and renders the animal less likely to excessive reflux as has been demonstrated in Chapter 5.

The decreased intraruminal pressure built up in a starved animal is due to the fact that if a sheep is starved for 24 hours, the ruminal ingesta are broken down by the microbes and little substrate is left for fermentation. This is why, less gas production occurs in the reticulorumen.

In the starved animals, the rumen contractions were very weak or absent in light anaesthesia. This is probably due to reduced volume of rumen contents and decreased rate of gas production which may not be able to excite the distension-sensitive receptors of the reticulorumen for vagal mediated reflex contractions (Leek, 1969). Another possibility is that, in the starved condition and during anaesthesia, the volatile fatty acids in the rumen may not be fully saturated and may pass through the gut in the unsaturated form which stimulates the secretion of cholecystokinin and this hormone is an inhibitor to gastric contractions (Nicholson and Omer, 1983).

It was presumed that nitrous oxide which tends to be accumulated in the body cavities (Hall and Clarke, 1983) might increase the intraruminal pressure and the pressure gradient, but these appeared to remain unchanged in starved animals in spite of continuous use of nitrous oxide throughout the period of anaesthesia lasting for 60 minutes. This suggests that accumulation of nitrous oxide in the reticulorumen if any, is insignificant particularly in the starved sheep.

Gastro-oesophageal reflux during anaesthesia did not appear to be directly related to the pressure gradient between oesophagus and reticulorumen since reflux was frequently not associated with maximum pressure gradient.

At rest the oesophagus is flaccid and its intraluminal pressure changes are transmitted from intrapleural pressures (Botha, 1959) which is usually negative. On the other hand, the reticulorumen always maintains a variable amount of positive pressure. This suggests the existence of some barrier at the gastro-oesophageal junction which prevents the transmission of positive intraruminal pressure towards the negative intra-oesophageal pressure. In the absence of such a barrier, the rumen contents must flow out into the oesophagus, particularly in a head down position. The nature of this barrier remains unclear. As previously stated, the barrier at the junction of the oesophagus and reticulorumen may be explained both anatomically and physiologically. For example, Thomas (1981) regarded this to be an anatomical barrier which included the intra-abdominal oesophagus, the right crus of the diaphragm, the gastric sling fibres and the redundant mucosa lining of the gastro-oesophageal junction. Ingelfinger (1958)

suggested that the gastro-oesophageal junctional zone has several mechanisms acting independently or in concert. These are (i) angulation of the oesophagus by diaphragmatic action, (ii) valvular structures created by the oblique entry of the oesophagus into the stomach, and (iii) intrinsic contraction or contractions of the gastro-oesophageal junctional area.

Conclusions

The pressure gradient between oesophagus and reticulorumen is not primarily involved in the initiation of reflux during anaesthesia. This gradient, however, is essential for the cranial movement of the rumen material.

There appears to be a regulatory mechanism at the junction of the oesophagus and reticulorumen which plays an important role in the occurrence of reflux. This regulatory mechanism would appear to have some of the properties of LOS as demonstrated in monogastric animals.

CHAPTER EIGHT

THE INFLUENCE OF INTRARUMINAL INSUFFLATION ON GASTRO-OESOPHAGEAL REFLUX

Introduction

As there is always a pressure gradient between oesophagus and reticulorumen, unless there is some form of "valve" at the junction of the reticulorumen and oesophagus, rumen contents would flow continuously into the oesophagus especially if the rumen is elevated above the oesophagus as in the head down position.

The lower oesophageal sphincter (LOS) is actively involved with the regulation of movement of ingesta between the oesophagus and reticulorumen along the pressure gradient. Observations in Chapter 7 suggested that the reflux of rumen contents during anaesthesia is not directly related to the gastro-oesophageal pressure gradient. Experiments were designed utilising gaseous insufflation of the rumen to allow manipulation of the intraruminal pressure and to test the efficacy of the LOS against controlled pressure gradient.

Materials and Methods

The technique of intraruminal insufflation involved preparation of the experimental animals with permanent ruminal cannulae.

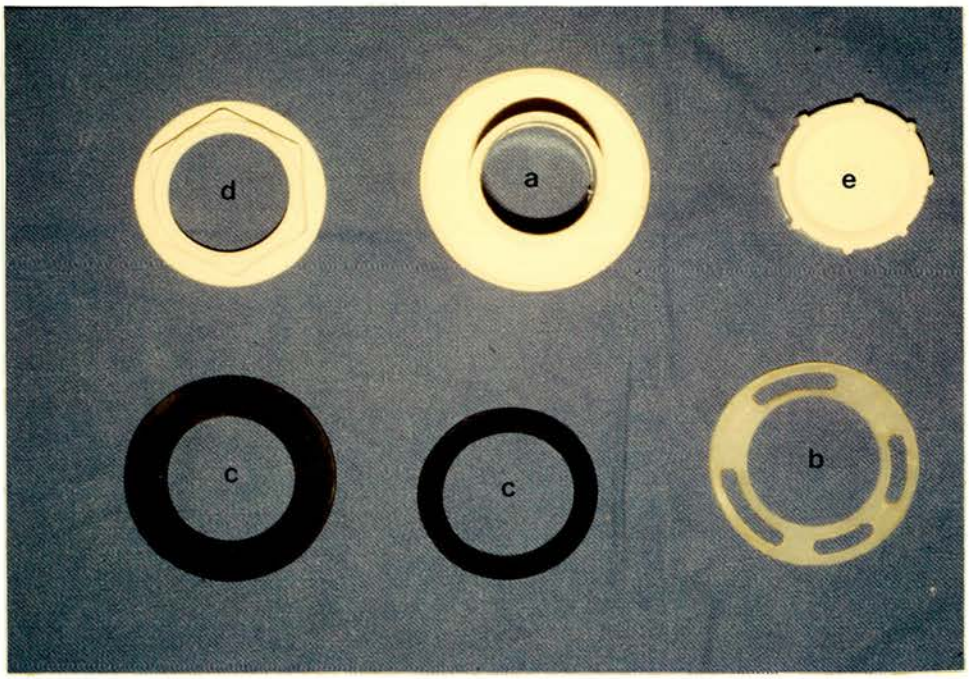
Ruminal Cannulation

Choice of cannulae materials: The cannula used in the present investigation was made of polyethylene vinyl chloride (PVC), the internal diameter being 4.1 cm. The intraruminal end was fitted with a PVC flange that ensured retention within the rumen. There were three separate rings, two were of rubber and one of PVC. Two PVC screws - one for tightening the cannula in position on the abdominal wall and the other functioning as an air-tight cap. The cannula is shown in Figure 8.1.

FIGURE 8.1

A. Polyvinyl chloride cannula for rumen fistula arranged to show individual parts. (a) shaft of the cannula; length of shaft 6.3 cm; width of collar at the base of the shaft 2.0 cm; internal diameter of the shaft 4.1 cm; (b) polyvinyl flange; width 1.5 cm; (c) rubber flanges; width 1.5 cm and 1.0 cm; (d) tightening screw; (e) air-tight screw top.

B. Cannula after assembly.


d
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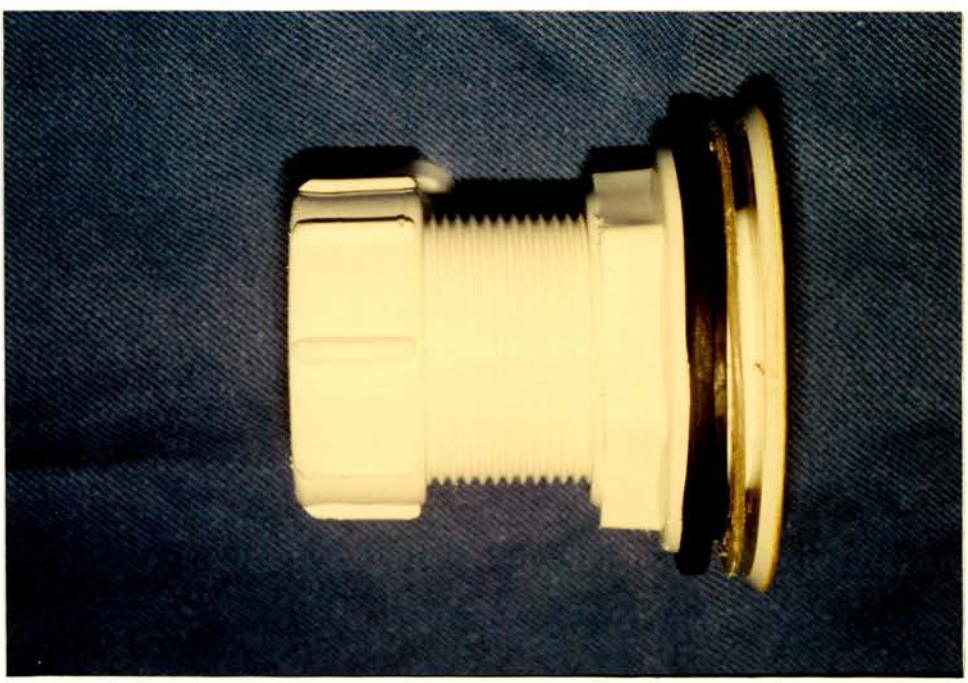


Preparation of the animal: Prior to operation, the animal was starved of food and water for 24 hours to reduce the bulk of the rumen as this has been demonstrated to minimise the risk of gastro-oesophageal reflux (Chapter 5).

Anaesthesia: Anaesthesia was induced by intravenous administration of alphaxolone/alphadolone (Saffan, Glaxovet Ltd.) at the dose rate of 3.0-6.0 mg kg^{-1} and maintained on a gaseous mixture of halothane, nitrous oxide and oxygen (50:50). The animal was positioned in right lateral recumbancy with the head tilted down.

Regional anatomy of the operative site: The site of operation was the left flank. The abdominal wall in this region is comprised of skin, the subcutaneous fascia, three layers of muscles and the parietal peritoneum. The blood supply to the abdominal muscles derives from the branches of lumbar arteries and from the terminal anterior branch of the deep circumflex iliac.

Operative technique: The operative site was surgically prepared by clipping, shaving and cleaning with povidone-iodine (Pevidine Surgical Scrub, Berk Pharmaceuticals Ltd., Shalford, Surrey, England). The shaved area was then sprayed with 0.5% chlorhexidine gluconate (Hibitane , ICI, Great Britain).

A modification of Dougherty's (1955) cannulation procedure was performed by using a single-stage technique. The position of the cannula on the left flank was land marked about 3.0-5.0 cm caudal to the last rib and as close to the transverse process as possible by putting a stab mark on the skin. A skin incision of adequate length (10.0-12.0 cm) and about 8.0 cm posterior to the land mark was made

vertically in the flank starting about 2.0 cm below the end of the transverse process of the third lumbar vertebra. The muscle layers were cut by blunt dissection and any haemorrhage was controlled by employing artery forceps. The peritoneum was severed and the rumen was located immediately below. A large pouch of the dorsal rumen was exteriorised and was held in place by tissue forceps. A purse-string suture using monofilament nylon was inserted into the pouch of the exposed rumen wall, penetrating all the coats except the mucosa. An incision was then made in the relatively avascular part in the centre of this area and was enlarged by scissors to allow the flange of the cannula to be introduced by slight stretching of the ruminal wound. The purse-string suture was then pulled tight and tied so as to invert the lips of the rumen wound. A second reinforcing purse-string suture was always applied over the first to ensure the firm positioning of the cannula. A PVC ring was sutured to the ruminal wall with monofilament nylon. A second incision was then made in the previously stab-marked location to allow the emergence of the cannula. The shaft of the cannula with screw cap was withdrawn through the stab opening as far as possible to ensure a close contact between the rumen and the parietal peritoneum. The two external rubber rings were put immediately above the skin and the first PVC screw was tightened over the outer rubber ring.

The muscle, subcutaneous fascia and skin of the initial wound were closed in layers by interrupted sutures using polyglycolic acid (Dexon, Cyanamid of Great Britain Ltd., Gosport, Hampshire). In order to prevent escape of the rumen contents over the peritoneum, the

cannula was always plugged with the PVC screw top. The major surgical steps are diagrammatically represented in Figure 8.2 and the completed operation is shown in Figure 8.3.

Post-operative maintenance: Animals with ruminal cannulae were kept in separate pens to minimise possible trauma to the cannulae which might lead to leakage of rumen contents into the surrounding tissues.

The body wall around the exterior flange of the cannula was regularly cleaned and the tension of the washers checked and adjusted to keep the cannula leak proof and to ensure that the tension was not enough to cause pressure necrosis.

A combination of procain penicillin and dihydro-streptomycin (Streptopen, Glaxovet Ltd.) was administered intramuscularly daily for the first four days post-operatively to reduce the risk of infection. The animals thus prepared were used for insufflation study starting two weeks after surgery.

Intraruminal Insufflation

Four unstarved sheep (including one Cheviot cross) with previously established rumen cannulae were used. Induction of anaesthesia was achieved with a mixture of halothane (4%), nitrous oxide and oxygen (50:50) using a face mask and Magill system (Chapter 4). Two planes of anaesthesia, light and deep, were maintained during this experiment. The selection of the first plane of anaesthesia was random. After induction, the screw top of the cannula was replaced by an air-tight rubber bung with a communicating polyethylene tube of internal diameter 6.0 mm. The rubber bung had also an extra perforation

FIGURE 8.2

Diagrammatic sketch of a single-stage rumen cannulation in sheep.

A. A pouch of section of the rumen is isolated, drawn through the body wall incision and is held with forceps, a purse-string suture employed on the exteriorized rumen pouch and a stab wound is made in the centre of the suture.

B. The flange of the cannula is inserted through the ruminal incision and the purse-string suture is tightened.

C. Cross section of the cannula in position. The edges of the ruminal wound were inverted as they were tightened around the cannula using purse-string suture.

D. Left flank of the sheep showing the site of operation and cannula positioned through a second stab incision in the flank between the initial wound and the last rib.

FIGURE 8.2

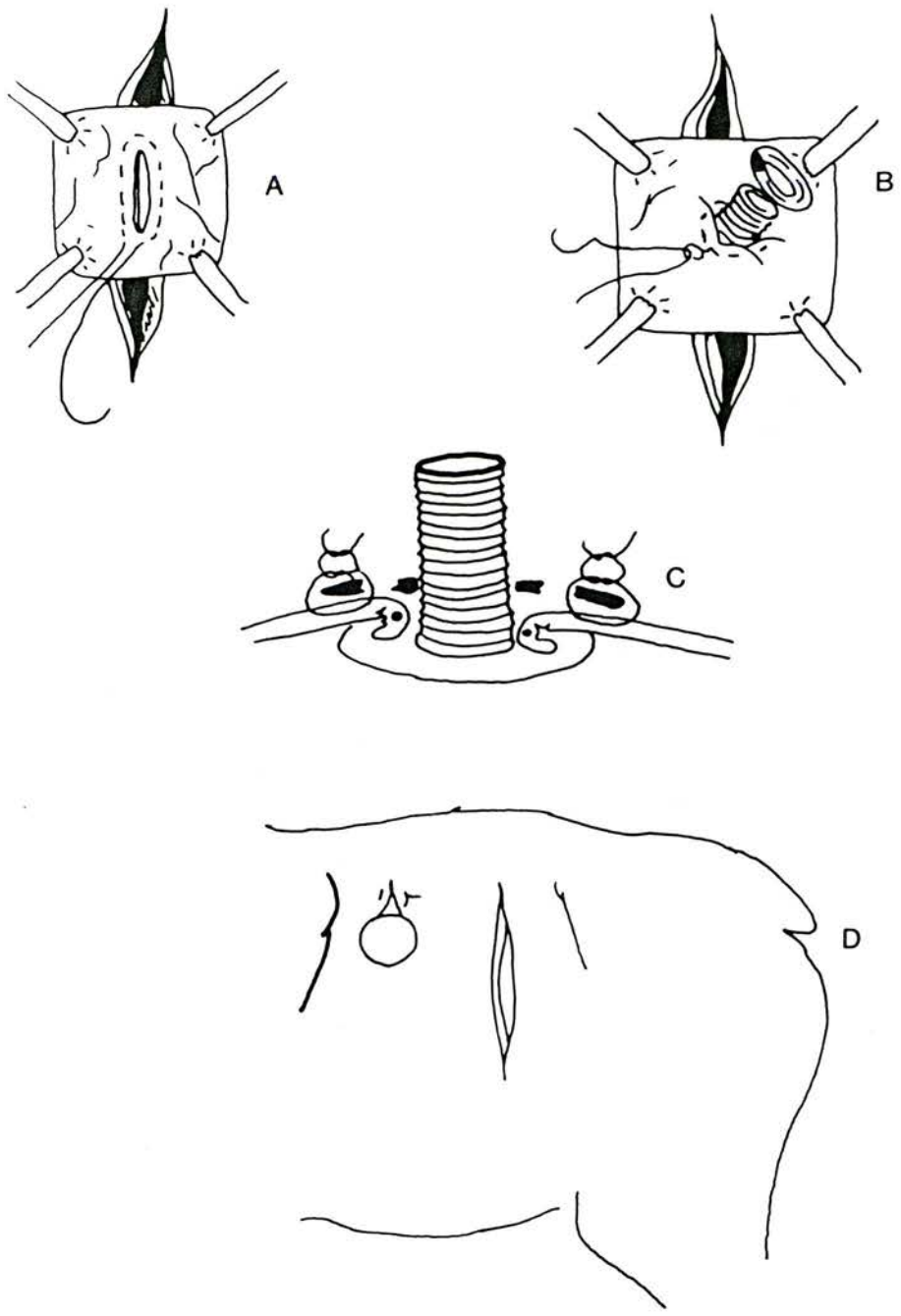


FIGURE 8.3

A sheep with rumen cannula.

to allow the entrance of a pressure transmitting catheter (outer diameter 2.0 mm) into the dorsal sac of the rumen.

For insufflation, the communicating tube was connected to a supply of oxygen from an anaesthetic machine. A flow meter was fitted to the system to enable the gas flow to be regulated.

In order to establish a convenient rate of gas flow, the rumen was insufflated with 3 litres, 5 litres and 10 litres per minute to produce reflux in either plane of anaesthesia. Oxygen was used as the insufflating gas throughout the experiment. For monitoring pressures during insufflation, the ruminal catheter was introduced through the cannula and the oesophageal catheter through the mouth. Insufflation was allowed to continue until reflux occurred. The presence of rumen contents, gas or both in the mouth was considered to be a reflux. Five minutes were allowed between insufflations and 10 insufflations were performed in each anaesthetic session. The experimental set up used for this study is illustrated in Figure 8.4.

The data obtained were analysed using the Student's t-test.

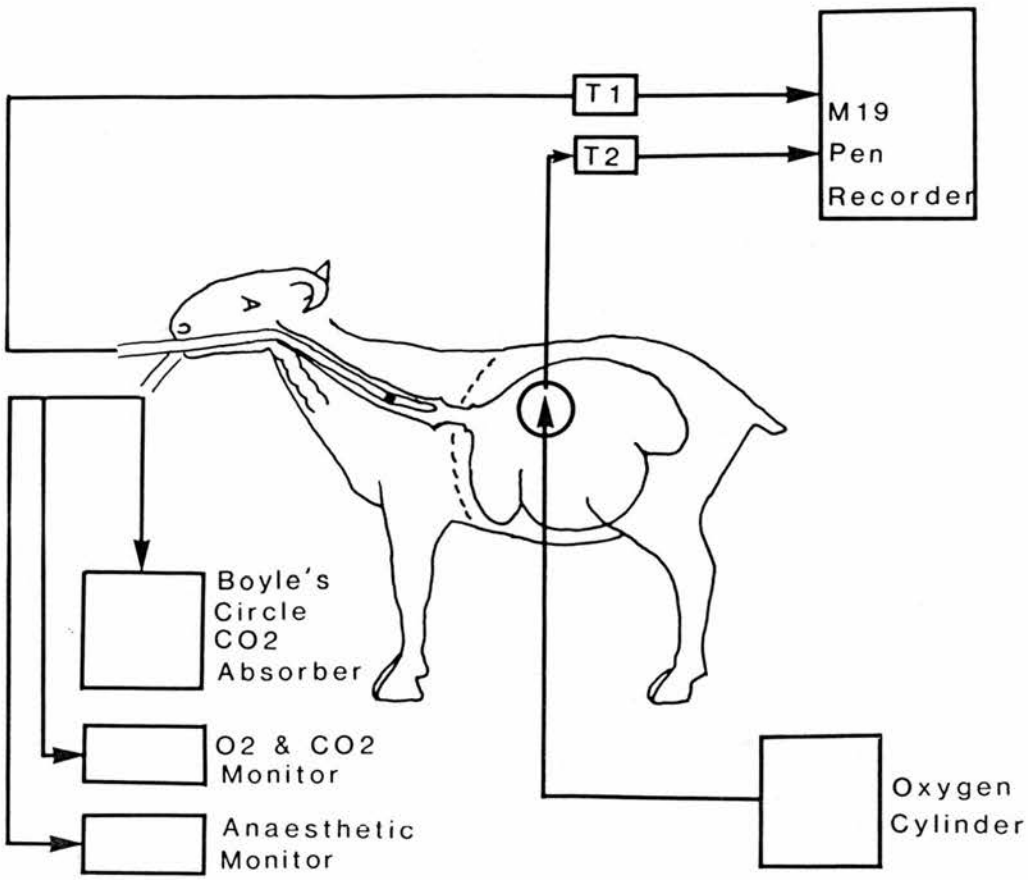
Results

The lower oesophageal sphincter (LOS) usually remained closed to the flow of rumen contents into the oesophagus during anaesthesia. When intraruminal pressure was increased by gaseous insufflation, the sphincter became patent and allowed the entrance of rumen contents or gas or both into the oesophagus.

In a control experiment, the effects of three different rates of intraruminal insufflation (3 l, 5 l and 10 l/minute) were studied in both light and deep planes of anaesthesia. Insufflation with 3 litres

FIGURE 8.4

EXPERIMENTAL SET-UP FOR INTRARUMINAL INSUFFLATION



T1,T2 : Pressure Transducers

or 5 litres per minute usually took a longer time to reach the threshold pressure required to produce LOS patency (Figures 8.5 and 8.6). With the increase of intraruminal pressure, breathing was seriously embarrassed and eventually ceased. This cessation of breathing made anaesthesia lighter and as a result, the effect of intraruminal insufflation in deep anaesthesia was not possible. The breathing however, was re-established as soon as the insufflation was stopped and the rumen was deflated.

Insufflation at 10 litres per minute was convenient in producing reflux in either plane of anaesthesia (Figures 8.7 and 8.8). At this rate of insufflation, the intraruminal pressure rapidly reached the threshold level to produce LOS patency and as a result, the depth of anaesthesia was not impaired, although cessation of breathing was unavoidable.

The effects of intraruminal insufflation on the occurrence of reflux during light and deep planes of anaesthesia are presented in Table 8.1. The intraruminal threshold pressure producing LOS patency in the light plane of anaesthesia was 39.64 ± 3.07 (S.D.) mm Hg and that in the deep plane was 33.69 ± 1.96 (S.D.) mm Hg. The difference was not statistically significant. Appendix 8.1 presents detailed records. Gaseous reflux was predominant and was usually associated with audible sound. The occurrence and nature of reflux during insufflation are shown in Table 8.2. Reflux of gas occurred in 62.5% of insufflations, rumen contents in 29.2% and gas and rumen contents in 8.3% of insufflations.

The increased intraruminal pressure due to insufflation interfered mechanically with respiration and was characterised by cessation

FIGURE 8.5

Intraruminal insufflation during light anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen; rate of insufflation - 3 litres per minute. The intraruminal pressure build up was very slow and commenced 3 minutes before this section of trace. The top horizontal bar indicates the length of insufflation. The thoracic oesophagus shows a series of pressure waves during insufflation without any visible reflux. The ruminal tracing presents weaker contractions. It was not safe to continue insufflation to produce reflux because of prolonged apnoea.

FIGURE 8.6

Intraruminal insufflation and the occurrence of reflux. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen; rate of insufflation - 5 litres per minute and commenced 2 minutes before this section of trace when the animal was in the deep plane of anaesthesia. The top horizontal bar indicates the length of insufflation. The thoracic oesophagus shows pressure waves associated with the occurrence of reflux. The ruminal tracing shows activity throughout the period of insufflation. Here also, it took a long time to produce a reflux and by this time the depth of anaesthesia became lighter due to prolonged suppression of inhalation of anaesthetic gases (note shallow respiratory excursions).

FIGURE 8.5

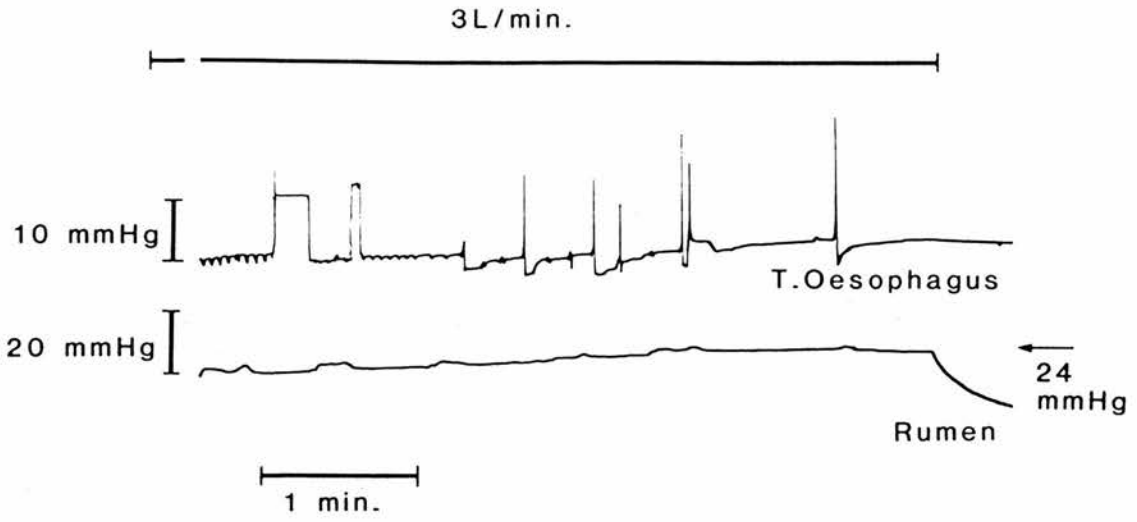


FIGURE 8.6

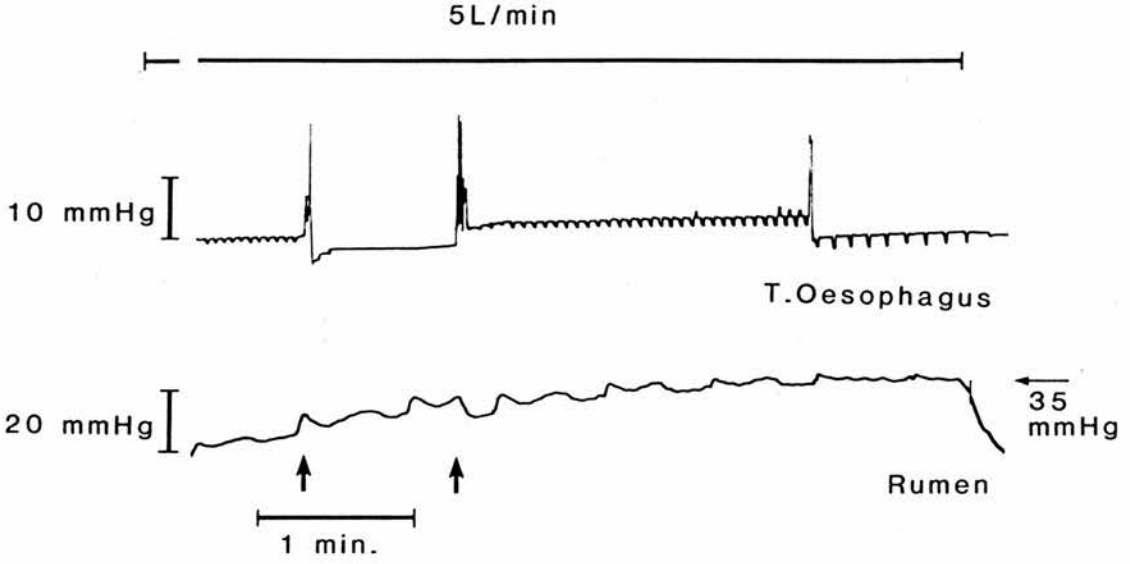


FIGURE 8.7

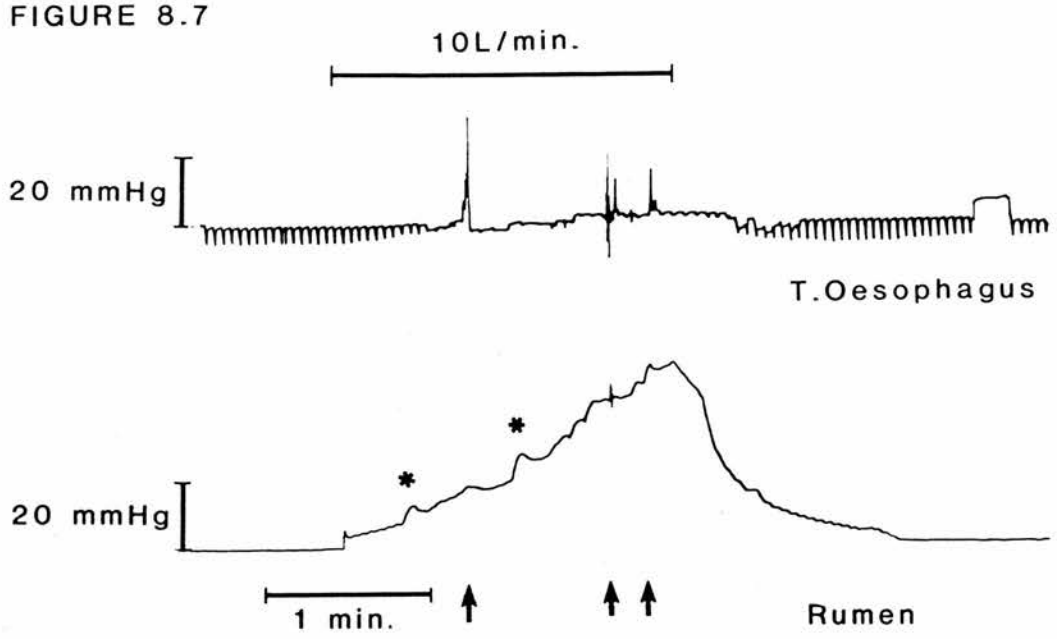


FIGURE 8.8

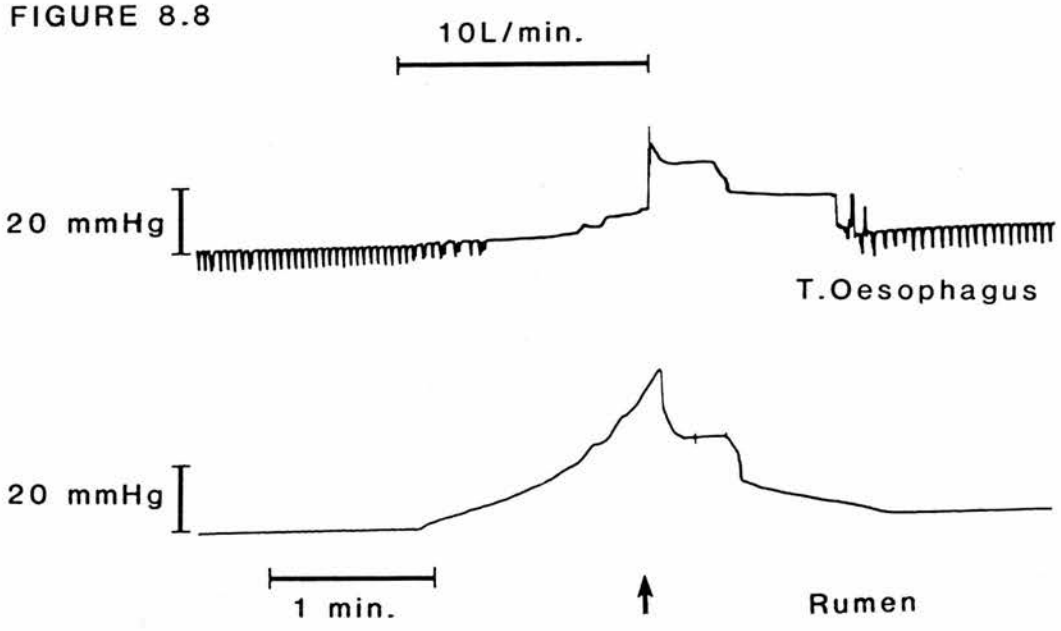


Table 8.1 Intraruminal insufflation and the pressure threshold producing LOS patency during light and deep planes of anaesthesia

Sheep Number	Number of anaesthetics	Intraruminal Threshold Pressure Producing LOS Patency (mm Hg)		P-value
		Light plane (n = 15)	Deep plane (n = 15)	
Black tag	3	42.13	36.13	> 0.05
230D	3	42.46	32.06	< 0.05
243D	3	37.4	34.46	> 0.05
27x91	3	36.6	32.13	> 0.05
Mean		39.64	33.69	> 0.05
S.D.		3.07	1.96	

Table 8.2 Intraruminal insufflation and the nature of reflux

Sheep Number	Number of insufflation	Nature of Reflux		
		Rumen gas	Rumen contents	Rumen gas + contents
Black tag	30	18	9	3
230D	30	21	8	1
243D	30	23	2	5
27x91	30	13	16	1
Total	120	75	35	10
%		62.5	29.2	8.3

of respiratory pressure changes in the oesophageal tracing (Figure 8.8). In a light plane of anaesthesia, the occurrence of reflux was always associated with some thoracic oesophageal activity (Figures 8.5-8.7). The rumen contractions were not interrupted during the course of insufflation in light anaesthesia. The occurrence of reflux in deep anaesthesia was not associated with thoracic oesophageal pressure waves (Figure 8.8). The rumen contractions were not normally present during a deep plane of anaesthesia.

Discussion

These results demonstrate the influence of controlled gastro-oesophageal pressure gradients on the occurrence of reflux by intraruminal insufflation. A successful intraruminal insufflation obviously depended on an efficient cannulation technique. The choice of material for the rumen cannula used in the present experiment was polyvinyl chloride (PVC) which is fairly resistant to the corrosion caused by the ruminal ingesta even on prolonged exposure. Earlier workers used cannulae made of ebonite (Quin, Van der Wath and Myburgh, 1938; Phillipson and Innes, 1939) and plastic or rubber and bakelite (Dougherty, 1955). Dougherty (1955) reported that bakelite and plastic cannulae may swell enough to cause the cork to stick. More recently, satisfactory rumen cannulation has been made with a polyethylene bottle fitted with a rubber compression plug (Komarek and Leffel, 1961).

In most cases after a variable delay, a small amount of rumen fluid leaks between the cannula and the scar ring which formed around the neck of the cannula, but the preparation is still able to sustain

increased intraruminal gas pressure. Ruminal cannulation does not seem to affect the digestive physiology and normal life expectancy, if healthy animals are used and when adequate precautions are taken (Dougherty, 1955). In the present experiment, all the cannulated animals were euthanized 6-9 months after surgery. The longest reported survival period in sheep with rumen cannula, however, is three years (Phillipson and Innes, 1939).

Cessation of breathing was a common feature associated with intraruminal insufflation. This is probably due to the progressive cranial compression of the diaphragm by the distended rumen. This supports the evidence by Musewe, Gillespie and Berry (1979) in cattle - "increased intraruminal insufflation (40 mm Hg) increases intrapleural and transdiaphragmatic pressure and a decreased lung volume". This alters the breathing pattern, inhibits mechanical activity of the diaphragm and finally induces respiratory failure (Musewe et al., 1979). This "breath holding" makes it difficult to maintain the period of deep anaesthesia when gaseous anaesthetics are employed. If the rate of insufflation is higher (10 litres/min), the threshold pressure for producing LOS patency is reached before the animal can become too light or experiences excessive respiratory embarrassment.

It has been suggested that the rumen may be insufflated with oxygen, nitrogen or with carbon-dioxide (Dougherty, Meredith and Barret, 1955). The intraruminal gas is absorbed from its wall (Kay, 1983). If the insufflation is with carbon-dioxide, its absorption from the rumen wall may further precipitate the already existing condition of respiratory acidosis. Insufflation with carbon-dioxide also

affects the rate of blood flow to the reticulorumen (Sellers, 1965) which may alter the motility of these organs. The use of oxygen for insufflation may help to protect against some of the adverse effects due to the impairment of respiration.

During the initial phase of insufflation, intraruminal pressure build up was accelerated (Figure 8.8). The reticulorumen is an elastic organ and with the increase of intraruminal pressure, the dimension of the organ is also increased up to a certain stage thereby the initial pressure build up is slow. As soon as the ruminal distension approaches a critical level, the gas pressure is rapidly increased.

In the present study, intraruminal insufflation with oxygen predominantly produced gaseous reflux. Intraruminal insufflation in conscious sheep has also been demonstrated to produce an increased rate of eructation (Dougherty, 1940; Weiss, 1953; Stevens and Sellers, 1959). During insufflation in light anaesthesia, the rumen motility increased and this observation is similar to that of Dougherty (1942) in conscious sheep. The occurrence of reflux during insufflation in light anaesthesia was associated with thoracic oesophageal pressure waves. These waves were probably due to peristaltic movement of the oesophagus which may have been stimulated by the hidden reflux of rumen material.

The pressure in the lower thoracic oesophagus was also passively increased on insufflation. This is due to the compression of the thoracic cavity by the distended rumen. No attempt was, however, made to monitor the responses of the LOS to intraruminal insufflation, since the position of the sphincter is progressively shifted cranially

with insufflation (Dodds et al., 1974) while that of the recording tip is fixed using the present technique.

It has been demonstrated in Chapter 7 that pressure gradient between the oesophagus and reticulorumen associated with reflux was 7.18 mm Hg. In the insufflation study, however, the intraruminal pressure required to produce reflux was 39.6 mm Hg in light anaesthesia. This pressure was unusually high in comparison to pressures that can be built up during normal periods of anaesthesia for 2-3 hours.

Conclusions

(i) Pressure gradient across the LOS is not the primary factor in initiating reflux. This gradient, however, may be important as the driving mechanism of reflux.

(ii) The factor that controls reflux is the "regulatory valve" which can be conveniently designated as the LOS.

CHAPTER NINE

A MANOMETRIC STUDY OF THE ACTIVITIES OF THE OESOPHAGUS AND RUMEN

Introduction

The reticulorumen and oesophagus and their sphincters are involved in the mechanism of reflux and regurgitation of rumen contents. Investigations in conscious and decerebrate preparations show that during regurgitation, the rumen contents are presented to the LOS by the active contractions of the reticulorumen. The rumen contents subsequently are carried to the oro-pharynx by active oesophageal antiperistalsis. Whether these activities of the oesophagus and reticulorumen are also involved during reflux in the anaesthetized condition have not previously been investigated. In this chapter, the physiological state of the oesophagus and rumen were studied during halothane anaesthesia with particular reference to gastro-oesophageal reflux. The influence of the plane of anaesthesia on the motor activities of the oesophagus and rumen were also investigated. Intraluminal manometry was utilised in this investigation. This technique has been employed extensively to study the motor activities of gastro-intestinal tract.

Materials and Methods

Eleven sheep were used in this investigation. Anaesthesia was induced with a mixture of halothane (4%), nitrous oxide and oxygen (50:50) and was maintained in a similar fashion as described in the general materials and methods (Chapter 4).

Oesophageal and ruminal activities were recorded using water filled open tip catheters (Catheter Assembly 2, Chapter 4). For simultaneous recording of pressures from these organs, the catheter assembly was positioned so that the oesophageal recording tip lay about

10.0 cm anterior and the ruminal recording tip about 10.0 cm posterior to the LOS. For determining the velocity of oesophageal pressure waves in light anaesthesia, the catheter assembly was occasionally withdrawn from the rumen to position both the recording tips along the thoracic oesophagus. In this position, it was possible to place the pressure recording tips in the thoracic oesophagus at a distance of 20.0 cm apart.

The response of the oesophagus to rumen material administered into the lower oesophagus was studied in three anaesthetics. In this case, the rumen material was collected from the same animal using a stomach tube. The collected rumen material (25 ml) was then injected into the thoracic oesophagus through a spare catheter using catheter assembly 2 (Chapter 4) while the pressure changes in the oesophagus were recorded.

The oesophageal response to balloon distension was studied using a separate catheter assembly as described in Chapter 4. The catheters were arranged so that a balloon could be distended with 25 cc air in either the thoracic or cervical oesophagus while the responses of the other parts of the oesophagus could be simultaneously monitored.

The data obtained in these studies were analysed by using the Student's t-test.

Results

Oesophageal and ruminal activities: These were studied in both light and deep planes of anaesthesia. The following events were monitored: gastro-oesophageal reflux, swallowing and "eructation".

The oesophageal activities were usually observed in the light plane of anaesthesia. The pressure waves of the thoracic oesophagus

associated with reflux in light and deep planes of anaesthesia are shown in Table 9.1. In light anaesthesia, in the majority of cases, reflux was associated with oesophageal activity, while in deep anaesthesia reflux never occurred in association with observed oesophageal activity.

Two patterns of oesophageal pressure waves were recorded: individual pressure waves (Figure 9.1) where the waves occurred singly and more or less in an isolated form, and groups of pressure waves (Figure 9.2) where two or more waves occurred together. The number of pressure waves in both planes of anaesthesia were analysed from 24 anaesthetics in four sheep (Table 9.2). The total number of pressure waves in the light plane of anaesthesia was 94 per hour as compared to only 4.5 per hour in the deep plane.

The plane of anaesthesia influenced the oesophageal and ruminal activity. In a light plane of anaesthesia, the rumen contractions were present in the majority of cases (Figure 9.3). The rate and amplitude of rumen contraction are presented in Table 9.3. The mean rate of rumen contraction was 0.7 per minute and the amplitude was 4.7 mm Hg. In this investigation, the rumen contractions were present in 83.3% of cases in the light plane of anaesthesia while in a deep plane, these contractions were absent in all cases (Table 9.4). As anaesthesia deepened, the amplitude of ruminal pressure waves progressively diminished and were finally abolished (Figure 9.4). In a deep plane of anaesthesia, it was unusual to get any pressure wave in either ruminal or oesophageal tracings other than the changes associated with respiration (Figure 9.5). The oesophageal tracing, however, was found to change its baseline occasionally in deep anaesthesia (Figure 9.6).

Table 9.1 The pressure waves of the thoracic oesophagus during reflux in periods of 30 minutes light and 30 minutes deep anaesthesia

Sheep No.	Number of anaesthetics	Number of reflux associated with oesophageal pressure waves		Number of reflux not associated with oesophageal pressure waves	
		Light plane	Deep plane	Light plane	Deep plane
192D	6	12	0	0	5
243D	6	1	0	0	7
27x91	6	0	0	0	1
289D	6	10	0	0	1
Total	24	23	0	0	14

Table 9.2 The pressure waves of the thoracic oesophagus in periods of 30 minutes light and 30 minutes deep anaesthesia

Sheep No.	Number of anaesthetics	Number of pressure groups		Total number of pressure waves including the groups	
		Light plane	Deep plane	Light plane	Deep plane
192D	6	72	0	575	0
243D	6	10	3	128	14
27x91	6	6	0	37	1
289D	6	44	10	395	39
Total	24	132	13	1135	54

N.B. The total number of pressure waves (including the groups) in light anaesthesia was not significantly higher than those in deep anaesthesia.

A group: More than one pressure wave at frequencies greater than 10 per minute.

Table 9.3 The rate and amplitude of rumen contractions in light plane of anaesthesia

Sheep No.	Number of recorded rumen contractions	Recorded time (mins)	Rate of rumen contraction per minute	Amplitude of rumen contraction (mm Hg)
192D	24	44	0.54	3.66
243D	69	98	0.70	3.76
27x91	77	104	0.74	5.87
289D	22	27	0.81	5.44
Mean			0.70	4.68
S.D.			0.11	1.13

Table 9.4 Influence of the plane of anaesthesia on rumen contractions

Sheep No.	Number of anaesthetics	Rumen Contractions			
		Light plane		Deep plane	
		Present	Absent	Present	Absent
192D	6	4	2	0	6
243D	6	6	0	0	6
27x91	6	5	1	0	6
289D	6	5	1	0	6
Total	24	20	4	0	24
%		83.3	16.7	0.0	100.0

FIGURE 9.1

Intraluminal pressure changes of the oesophagus and rumen during light anaesthesia using open tip catheter (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The oesophageal tracing shows typical independent singular pressure waves (superimposed on smaller respiratory pressure changes).

FIGURE 9.2

Intraluminal pressure changes of the oesophagus and rumen during light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The oesophageal tracing shows groups (a group is defined as two or more pressure waves occurring at intervals of less than 10 seconds) of pressure waves. The ruminal tracing shows some weak pressure waves but do not correspond to oesophageal pressure waves.

FIGURE 9.1

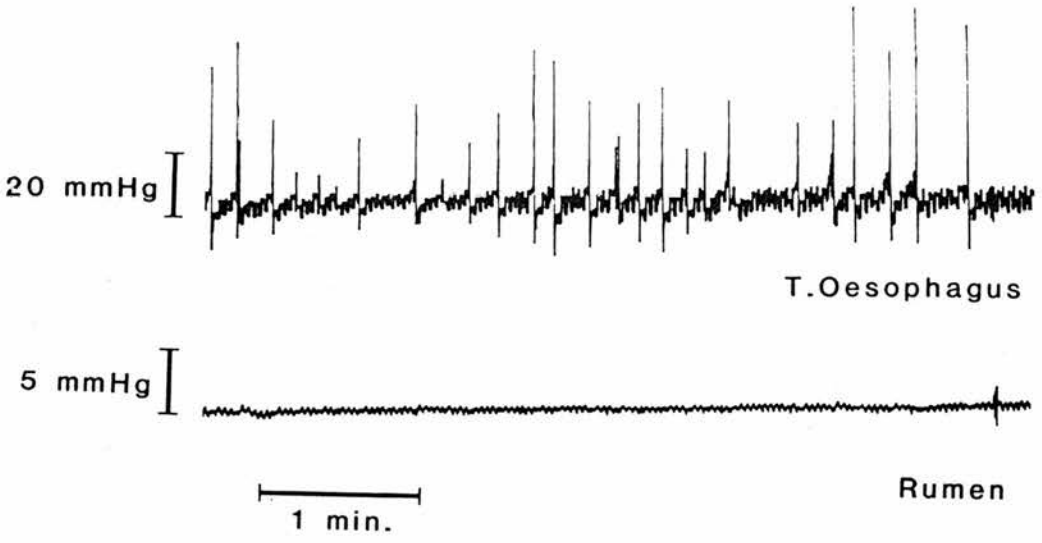


FIGURE 9.2

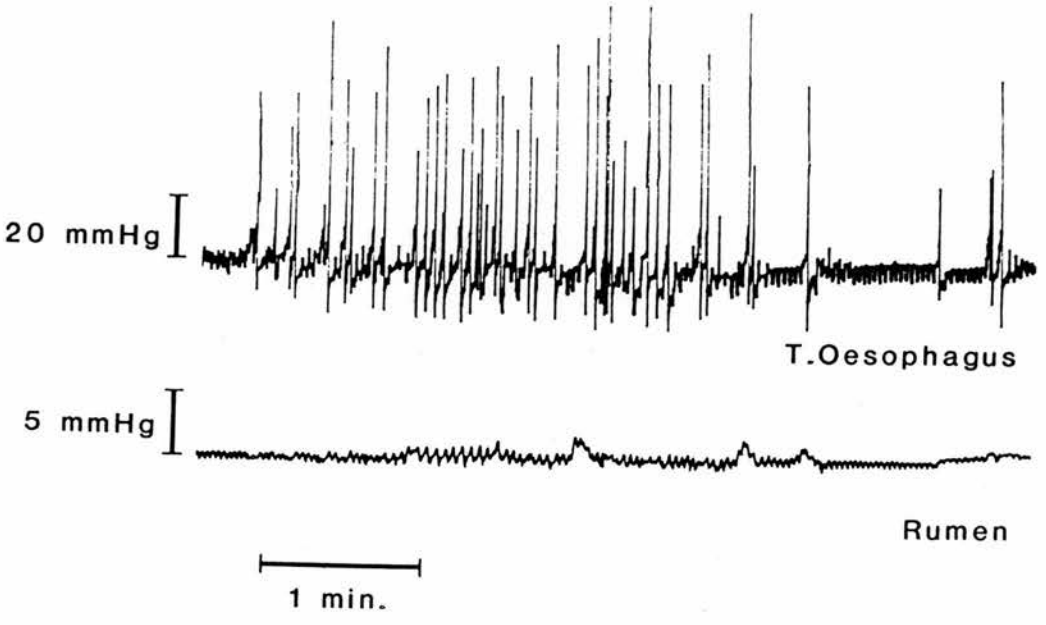


FIGURE 9.3

Intraluminal pressure changes of the oesophagus and rumen during light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The ruminal pressure waves occurred at an approximate frequency of one per minute. The oesophageal tracing shows pressure changes associated with respiration.

FIGURE 9.4

Intraluminal pressure changes of the oesophagus and rumen as anaesthesia deepened (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The amplitudes of ruminal pressure waves were progressively diminished and eventually abolished with the increase of the depth of anaesthesia. The arrow indicates the point when the concentration of halothane in expired gases was increased between 1.5-2.0%.
At the second arrow pointing to the thoracic oesophagus, the paper speed of the recorder was increased (100 mm/minute) to see the individual pressure wave associated with breathing.

FIGURE 9.3

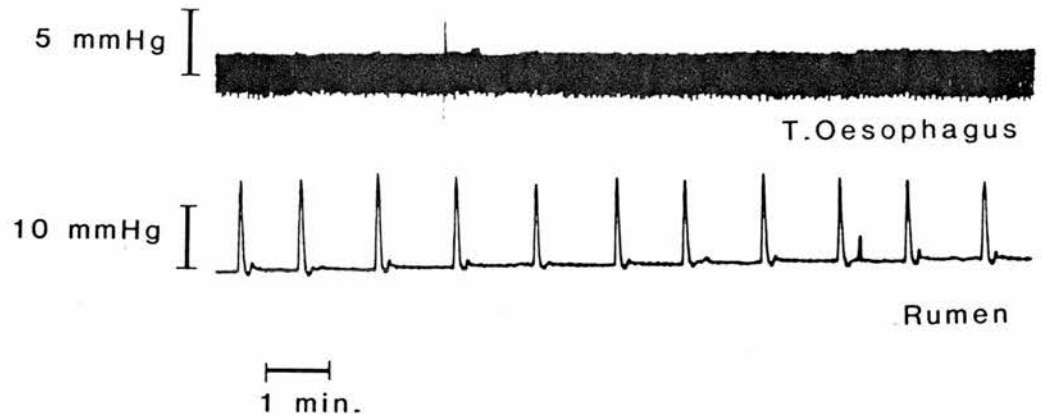


FIGURE 9.4

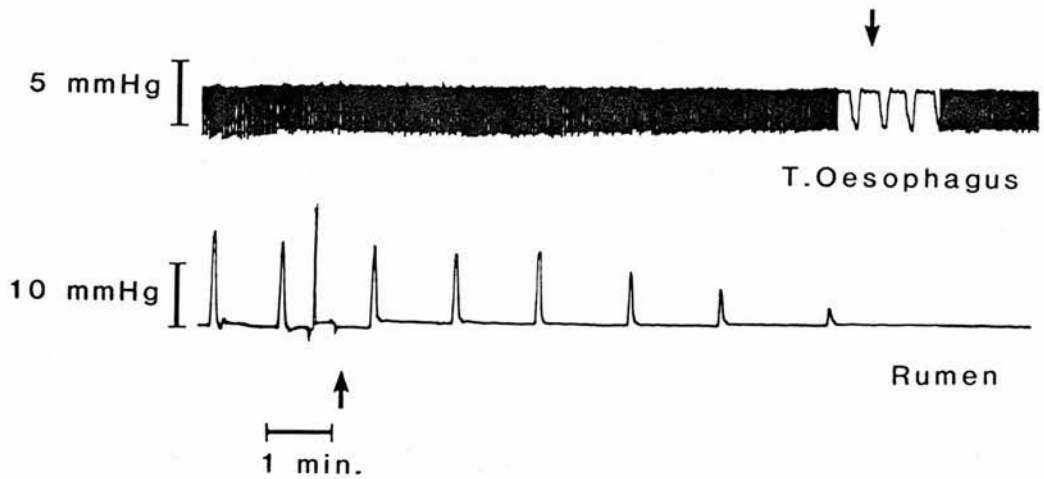


FIGURE 9.5

Intraluminal pressure changes of the oesophagus and rumen during deep anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). During deep anaesthesia, neither the thoracic oesophagus nor rumen showed any additional pressure wave apart from changes associated with breathing.

FIGURE 9.6

Intraluminal pressure changes of the oesophagus and rumen during deep anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The oesophageal tracing showed an abrupt increase in resting tone while the ruminal tracing showed a gradual build up of intraruminal pressure.

FIGURE 9.5

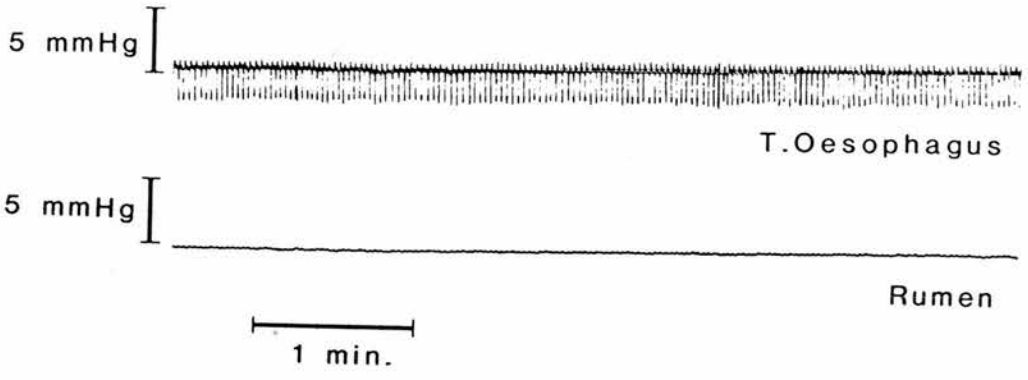
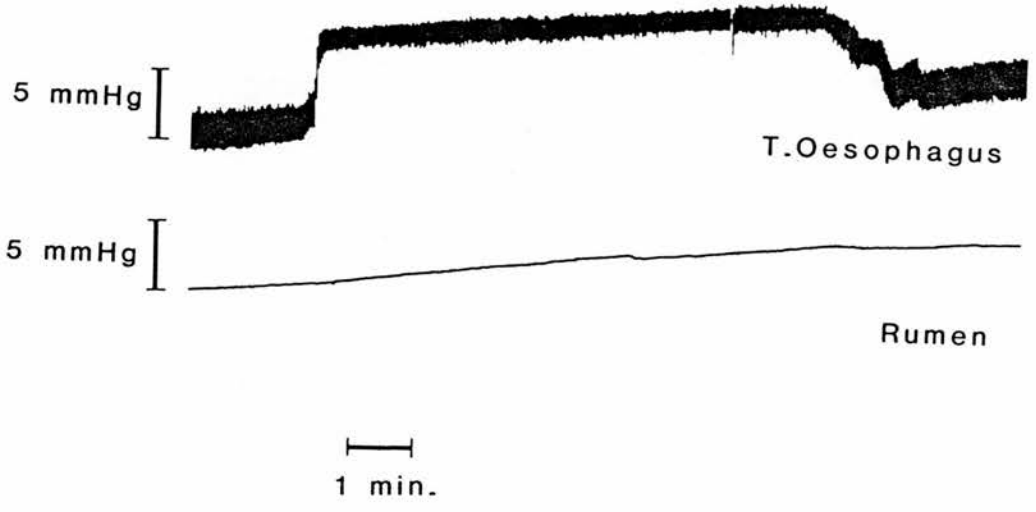


FIGURE 9.6



As anaesthesia lightened, the abolished ruminal pressure waves re-appeared with a progressive increase in amplitude (Figure 9.7). There was usually no temporal correlation between the pressure waves of the thoracic oesophagus and those of the rumen. On a few occasions, however, these pressure waves were found to precede the rumen contractions (Figure 9.8).

During light planes of anaesthesia when the oesophagus was undergoing pressure changes, the catheter assembly was withdrawn from the rumen and fixed at a position so that the terminal recording tip lay in the posterior thoracic oesophagus and the proximal one in the anterior thoracic oesophagus to monitor the direction of these pressure waves. These pressure waves were consistently found to be peristaltic, i.e. the direction of the waves was aboral (Figure 9.9). These activities were not associated with any visible swallowing, "eructation" or reflux. Frequently, these pressure waves were confined to the caudal part of the thoracic oesophagus (Figure 9.10).

The pattern of oesophageal pressure waves during reflux was also recorded and these were again consistently peristaltic in nature (Figure 9.11).

The duration, amplitude and velocity of the peristaltic contractions varied at different levels of the thoracic oesophagus. The mean duration, amplitude and velocity of these pressure waves during reflux are presented in Table 9.5. The duration and amplitude of the oesophageal pressure waves in the anterior thoracic oesophagus were 1.12 ± 0.27 (S.D.) seconds and 11.54 ± 1.81 (S.D.) mm Hg and those in the posterior thoracic oesophagus were 1.47 ± 0.25 (S.D.) seconds and 8.94 ± 2.13 (S.D.) mm Hg. These differences were statistically significant ($P < 0.05$). Appendix 9.1 presents detailed records.

Table 9.5 Duration, amplitude and velocity of thoracic oesophageal pressure waves associated with reflux in the light plane of anaesthesia

Sheep No.	Thoracic Oesophageal Pressure Waves				Wave velocity (cm/sec)
	Anterior thoracic oesophagus		Posterior thoracic oesophagus		
	Duration (sec) n = 25	Amplitude (mm Hg) n = 25	Duration (sec) n = 25	Amplitude (mm Hg) n = 25	
271D	0.80	9.68	1.18	8.54	25.86
127B	1.34	11.76	1.45	7.58	25.59
518E	1.32	12.60	1.90	8.82	27.26
Z981	1.02	10.28	1.62	7.96	23.93
645B	0.83	10.42	1.41	13.18	27.33
566E	1.41	14.54	1.30	7.58	36.99
Mean	1.12	11.54	1.47	8.94	28.87
S.D.	0.27	1.81	0.25	2.13	5.63

Significance of differences between anterior and posterior thoracic oesophageal pressure waves:
Duration $P < 0.05$
Amplitude $P < 0.05$

FIGURE 9.7

Intraluminal pressure changes of the oesophagus and rumen as anaesthesia lightened (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The ruminal pressure waves reappeared with progressive increase in amplitude with the decrease in depth of anaesthesia. The arrow indicates a point when the concentration of halothane in expired gases was brought down between 0.5-1.0%.

FIGURE 9.8

Intraluminal pressure changes of the oesophagus and rumen during light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Strong oesophageal pressure waves occurred preceding ruminal pressure waves.

FIGURE 9.7

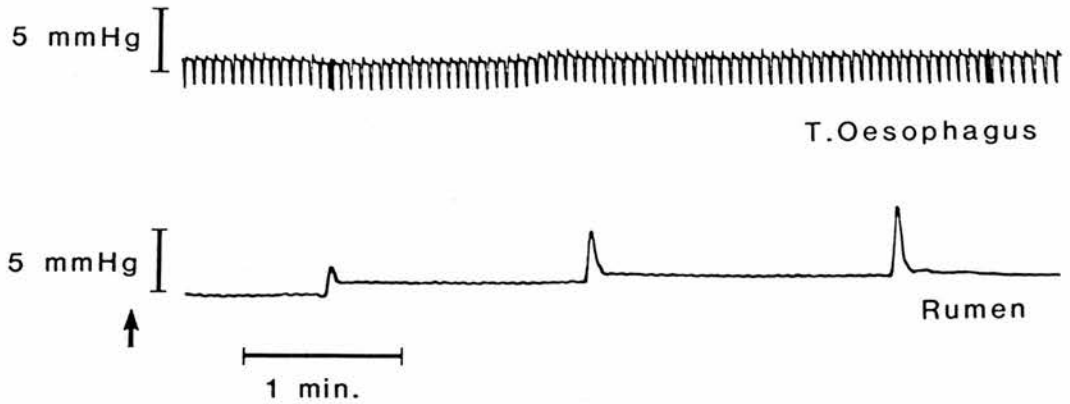


FIGURE 9.8

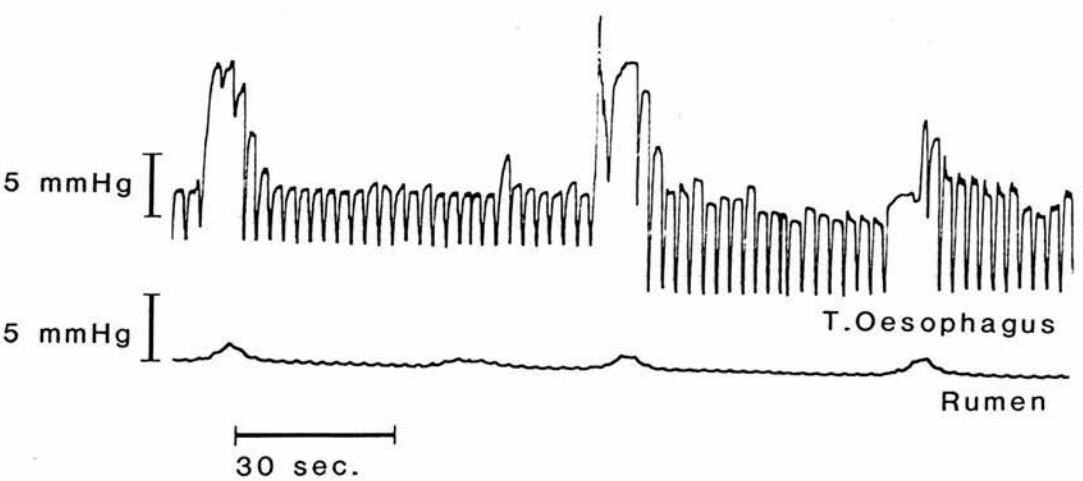


FIGURE 9.9

Direction of the oesophageal pressure waves during light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves of the anterior thoracic oesophagus preceded those of posterior thoracic oesophagus demonstrating that these waves were peristaltic. The velocity of these pressure waves can be calculated from the following equation:-

$$V = \frac{d}{t}$$

where V = velocity

d = distance between the two recording tips (in cm)

t = time taken by a pressure wave to travel the distance (in sec)

The velocity is expressed in terms of $cm.sec^{-1}$

FIGURE 9.10

Activities of the thoracic oesophagus during light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves were more frequently present in the posterior thoracic oesophagus in contrast to anterior thoracic oesophagus.

FIGURE 9.9

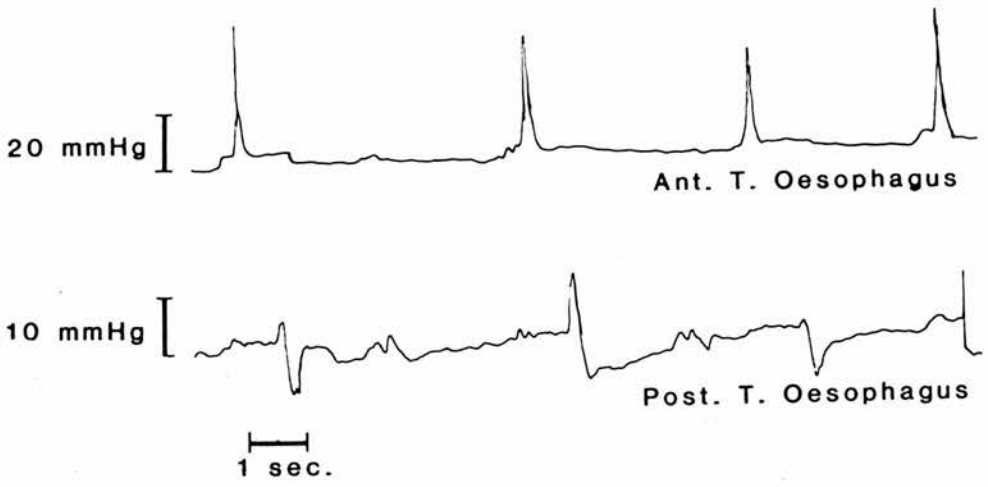


FIGURE 9.10

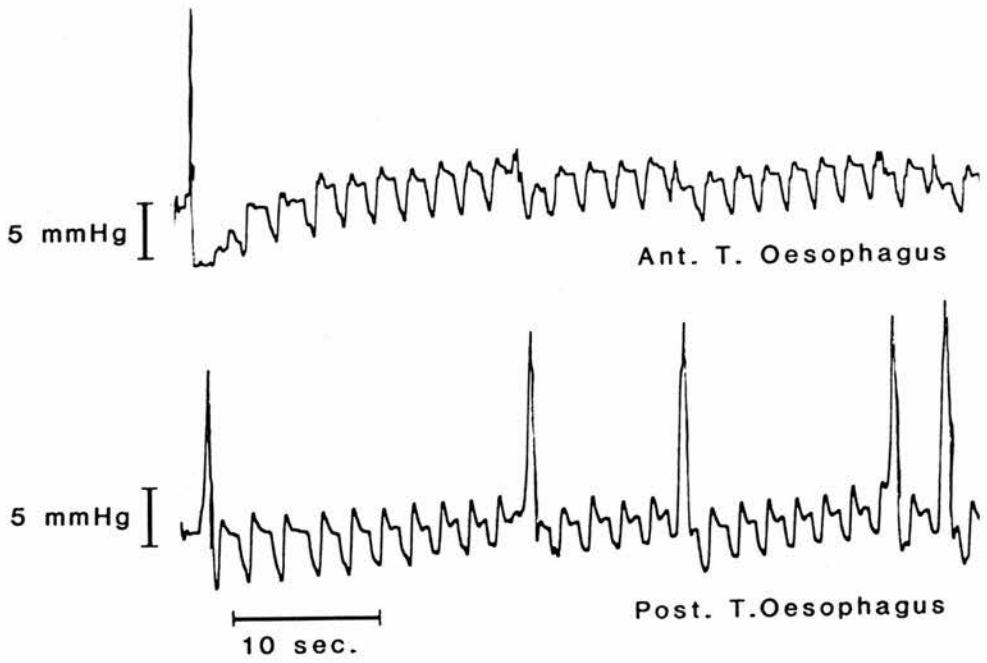


FIGURE 9.11

Direction of the oesophageal pressure waves during continuous reflux in light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves of the anterior thoracic oesophagus preceded those of posterior thoracic oesophagus demonstrating that these waves were peristaltic.

FIGURE 9.12

Duration of the oesophageal pressure waves during light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves of the posterior thoracic oesophagus had longer duration than those in the anterior thoracic oesophagus.

FIGURE 9.11

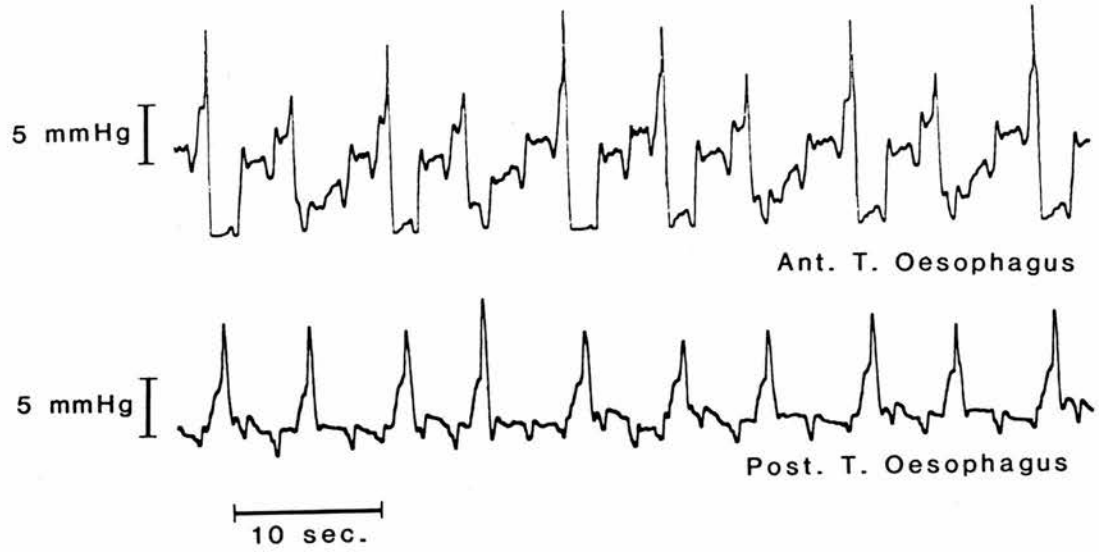
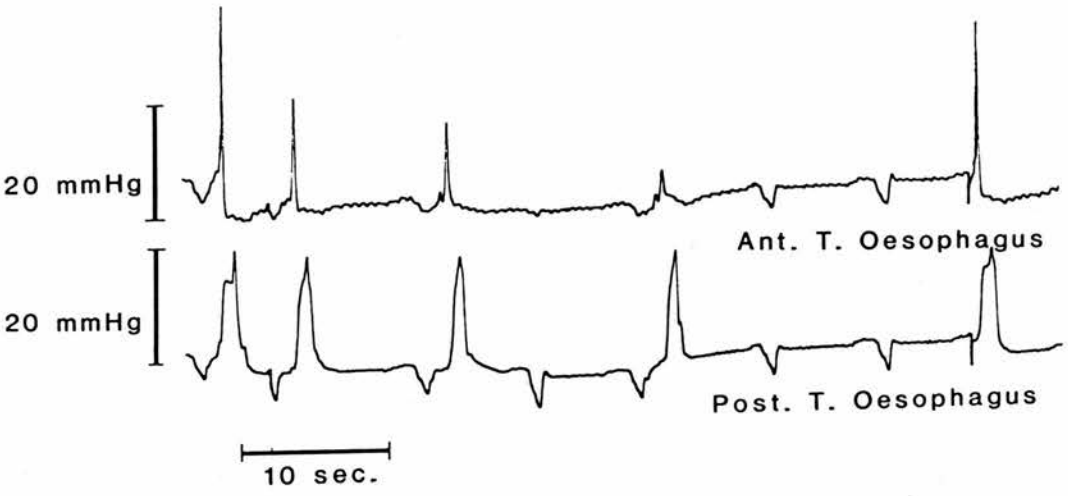


FIGURE 9.12



The mean duration, amplitude and velocity of the thoracic oesophageal pressure waves not associated with reflux in light anaesthesia are presented in Table 9.6. The duration of pressure waves in the anterior thoracic oesophagus was 0.85 ± 0.18 (S.D.) seconds and the amplitude was 11.43 ± 4.04 (S.D.) mm Hg whereas in the posterior thoracic oesophagus, the duration was 1.41 ± 0.26 (S.D.) seconds and the amplitude was 15.22 ± 2.29 (S.D.) mm Hg. The difference in duration was highly significant ($P < 0.001$). The individual records are presented in Appendix 9.2.

To summarise these results, the mean duration of oesophageal pressure waves in the posterior thoracic oesophagus was always (either associated or not associated with reflux) significantly greater than that in the anterior thoracic oesophagus. There was, however, no significant difference in amplitude between these groups.

The duration of the pressure waves in the anterior and posterior thoracic oesophagus is shown in Figure 9.12. During anaesthesia, the cervical oesophagus usually showed relatively less activity in comparison with thoracic oesophagus (Figure 9.13). The intraluminal pressure changes of thoracic oesophagus due to respiration at different levels were different during anaesthesia (Figure 9.14). The amplitude of these pressure waves was greater in the posterior thoracic oesophagus than the anterior.

When the depth of anaesthesia lightened sufficiently, visible swallowing movements were found to occur. The pressure changes associated with swallowing are shown in Figure 9.15. Prior to swallowing, there was a gradual pressure build up in the thoracic oesophagus, then a sharp pressure wave followed by a sudden drop below the resting base-

Table 9.6 Duration, amplitude and velocity of thoracic oesophageal pressure waves not associated with reflux in the light plane of anaesthesia

Sheep No.	Thoracic Oesophageal Pressure Waves				Wave velocity (cm/sec)
	Anterior thoracic oesophagus		Posterior thoracic oesophagus		
	Duration (sec) n = 25	Amplitude (mm Hg) n = 25	Duration (sec) n = 25	Amplitude (mm Hg) n = 25	
Z981	1.08	15.36	1.64	16.14	25.46
127B	0.86	10.02	1.37	10.52	25.17
518E	0.82	9.88	1.82	15.28	28.59
566E	1.13	6.12	1.38	17.44	25.83
645B	0.68	10.66	1.50	14.36	28.14
Z650	0.68	18.24	1.08	16.26	25.06
Mean	0.85	11.43	1.41	15.22	26.37
S.D.	0.18	4.04	0.26	2.29	1.57

Significance of differences between anterior and posterior thoracic oesophageal pressure waves:

Duration $P < 0.001$
Amplitude $P > 0.05$

FIGURE 9.13

Activities of the cervical and thoracic oesophagus during light anaesthesia (OTC). Upper tracing - cervical oesophagus (10.0 cm anterior to thoracic inlet); lower tracing - thoracic oesophagus (10.0 cm posterior to thoracic inlet). The cervical oesophagus showed very little activity as compared to the thoracic oesophagus.

FIGURE 9.14

Pressure changes in the anterior and posterior thoracic oesophagus during anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus; lower tracing - posterior thoracic oesophagus. The amplitude of the respiratory pressure changes was usually greater in the posterior thoracic oesophagus than the anterior.

FIGURE 9.13

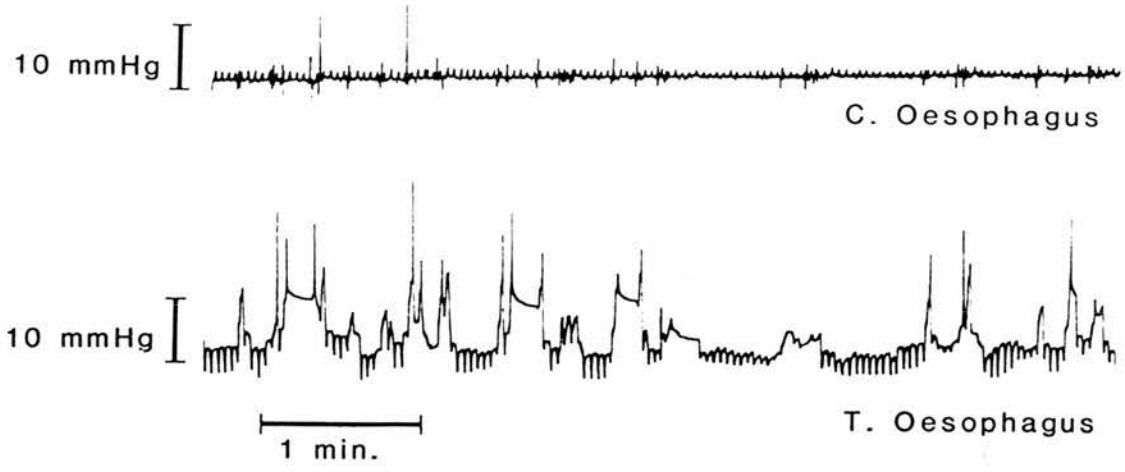


FIGURE 9.14

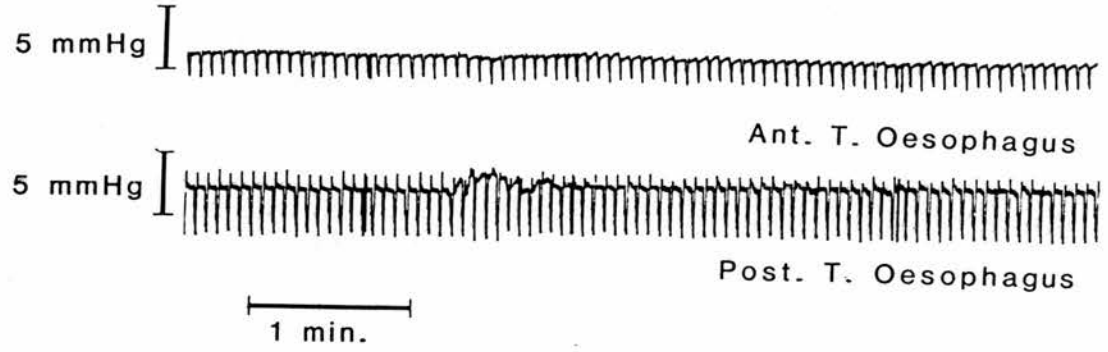


FIGURE 9.15

Intraluminal pressure changes of the thoracic oesophagus and rumen associated with swallowing (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrow indicates the onset of a swallow. The thoracic oesophagus showed an increase in tone prior to and a spike at the onset of a swallowing followed by a sudden fall in tone below the resting level associated with a series of pressure waves. The ruminal pressure waves did not associate with the swallowing activities.

FIGURE 9.16

Intraluminal pressure changes of the thoracic oesophagus and rumen during gaseous reflux in light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the timing of occurrence of observed gaseous reflux. The ruminal pressure waves were consistently present during gaseous reflux and were associated with a series of thoracic oesophageal pressure waves.

FIGURE 9.15

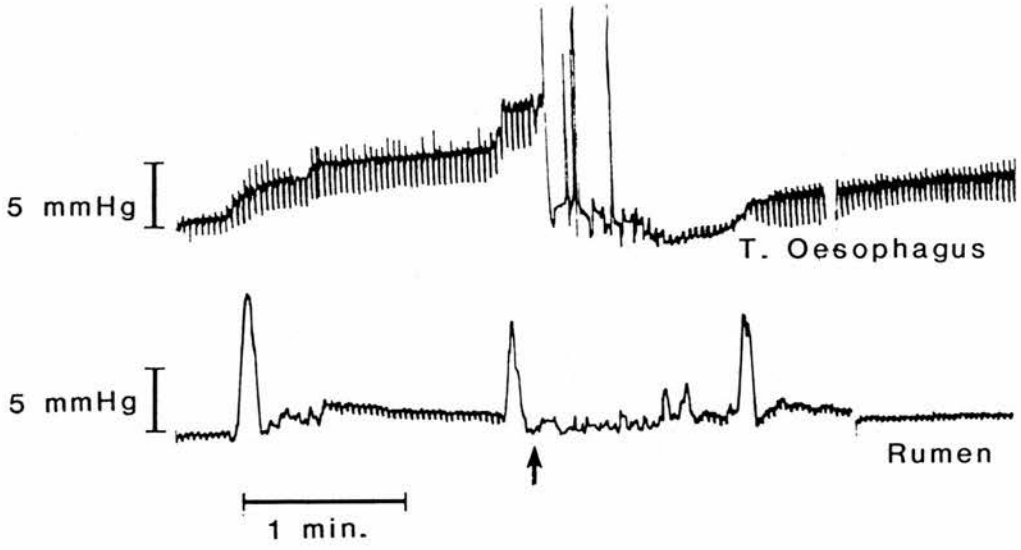
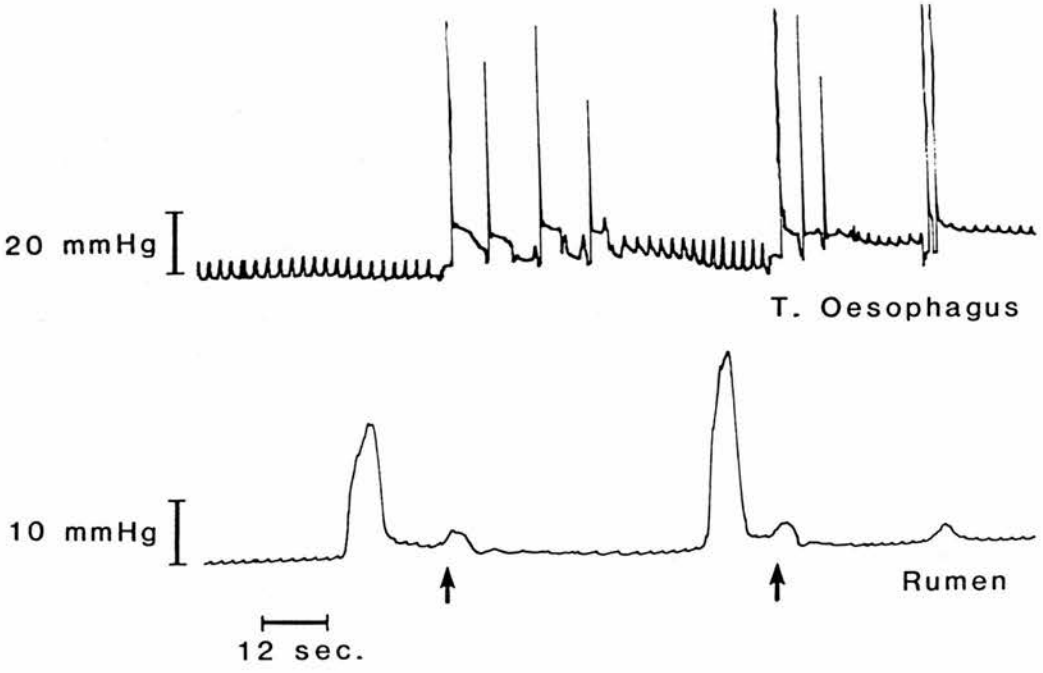


FIGURE 9.16

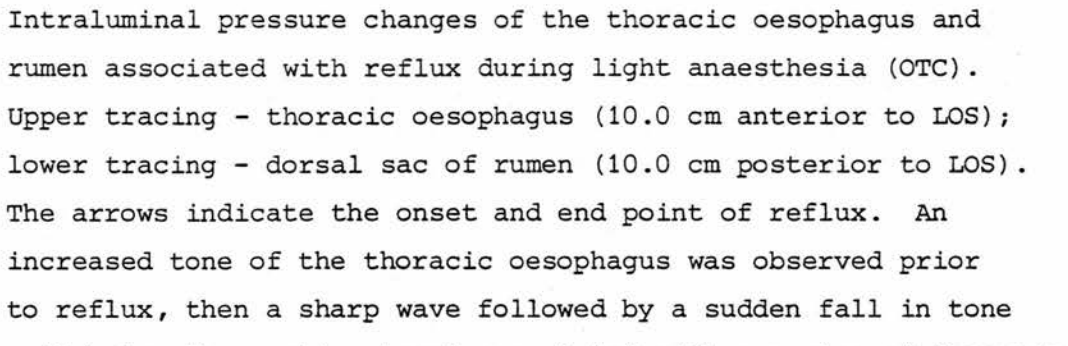


line pressure and was associated with a series of thoracic oesophageal pressure waves.

The occurrence of gaseous reflux during anaesthesia whether light or deep was unusual and the expulsion of rumen gas through the mouth was observed in only one anaesthetic. The ruminal and oesophageal pressure changes associated with this event were recorded (Figure 9.16). As can be seen from these tracings, the rumen appears to contract twice, the first of these being considerably more forceful than the second. Following the second and smaller contraction, a series of typical oesophageal pressure waves were recorded. Expulsion of ruminal gas was detected visibly and audibly travelling towards the mouth. This appeared to be related to the second ruminal contraction. Since there were not two recording tips in the oesophagus, it was not possible to record the direction of these pressure waves. Visual observation of swallowing in the pharynx and cervical oesophagus indicates that at this level these waves were peristaltic in nature. This is supported by the observation that these waves occurred after the reflux of gas and after the first swallowing movement in the pharynx and cervical oesophagus.

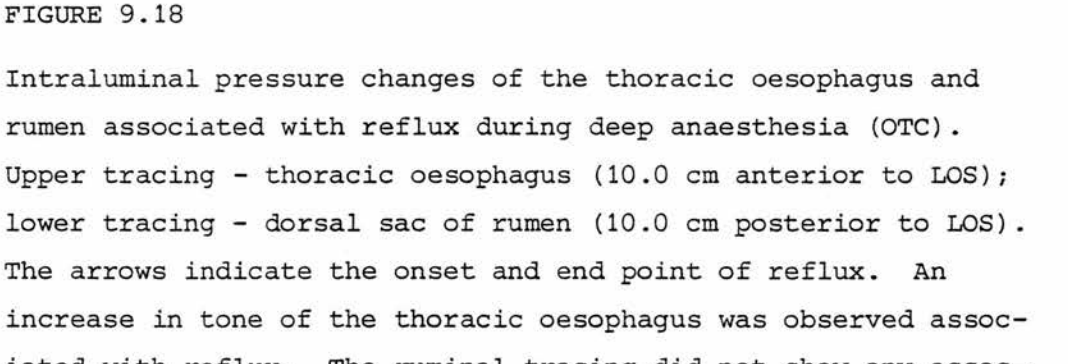
The pressure changes in the thoracic oesophagus associated with reflux in light anaesthesia are presented in Figure 9.17. Prior to reflux, the tone of the thoracic oesophagus was increased, then a sharp pressure wave was followed by a sudden drop well below the resting baseline pressure associated with a series of thoracic oesophageal pressure waves. The ruminal tracing, on the other hand, exhibited irregular and weak pressure waves regardless of the onset of reflux.

FIGURE 9.17



Intraluminal pressure changes of the thoracic oesophagus and rumen associated with reflux during light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the onset and end point of reflux. An increased tone of the thoracic oesophagus was observed prior to reflux, then a sharp wave followed by a sudden fall in tone well below the resting level associated with a series of thoracic oesophageal pressure waves. The rumen did not show any corresponding change.

FIGURE 9.18



Intraluminal pressure changes of the thoracic oesophagus and rumen associated with reflux during deep anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the onset and end point of reflux. An increase in tone of the thoracic oesophagus was observed associated with reflux. The ruminal tracing did not show any associated change.

FIGURE 9.17

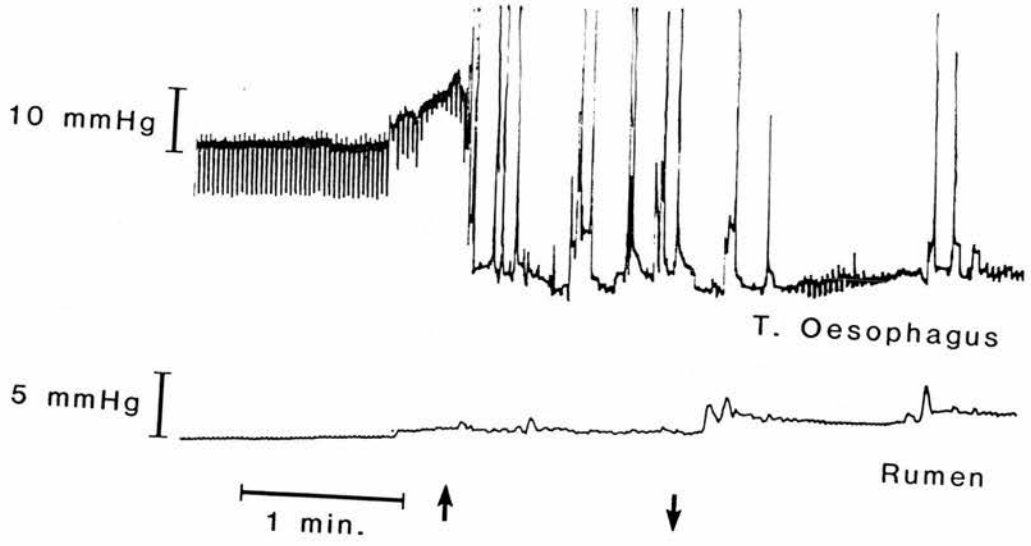
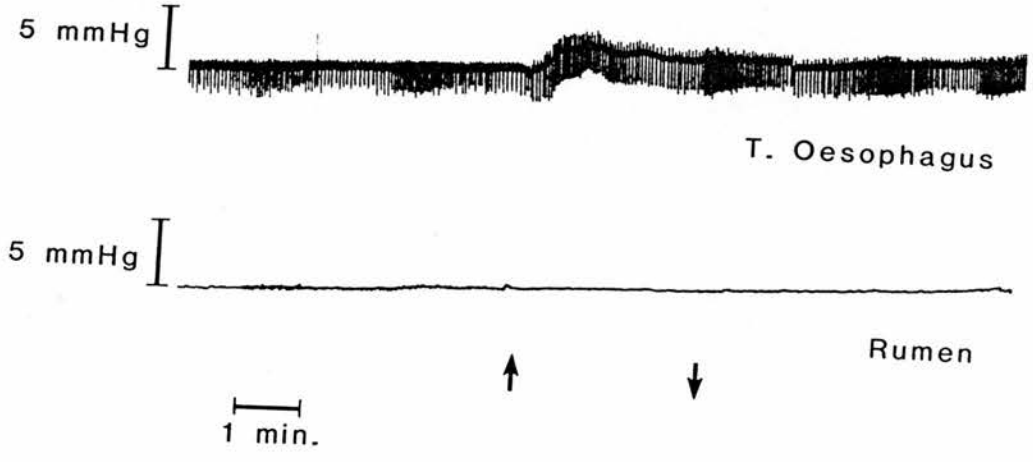


FIGURE 9.18



Reflux during deep anaesthesia was not usually associated with ruminal or oesophageal activity. Occasionally, the thoracic oesophageal tone was increased at reflux and this increased tone was maintained for some time before dropping to the baseline (Figure 9.18).

The oesophageal pressure wave usually remained unaffected by the pressure changes in the rumen. In one anaesthetic, however, the pressure waves due to rumen contractions were transmitted to the thoracic oesophagus (Figure 9.19).

Oesophageal response to injected rumen fluid: The pressure changes in the thoracic oesophagus in response to injection of 25 ml rumen fluid in light anaesthesia are shown in Figure 9.20. The thoracic oesophagus manifested peristaltic movements in one sheep. This response, however, was not consistent and no oesophageal peristalsis occurred in the other two sheep.

Oesophageal response to balloon distension: The responses of the thoracic oesophagus to balloon distension (4.0 cm long balloon with 25 cc air) were recorded. The responses of the oesophagus to intraluminal balloon distension are presented in Appendices 9.3 and 9.4. When a balloon was inflated in the cervical oesophagus in light anaesthesia, the tone (a maintained change in baseline pressure lasting for periods of greater than one minute) of the anterior thoracic oesophagus was increased in 71.4% of cases, decreased in 9.5% of cases and 19% did not produce any change, while in deep anaesthesia, these figures were 50%, 0.0% and 50% respectively. The responses of posterior thoracic oesophagus to such balloon distension was also monitored. The tone was increased in 66.7% of cases, decreased in 0.0%

FIGURE 9.19

Intraluminal pressure changes of the thoracic oesophagus in anaesthetized sheep with incompetent LOS during continuous reflux (OTC). Upper tracing - anterior thoracic oesophagus; lower tracing - posterior thoracic oesophagus. The increased intraruminal pressures during rumen contractions were transmitted into the thoracic oesophagus in a regular fashion. The pressure waves in the posterior thoracic oesophagus preceded those of anterior thoracic oesophagus, because the recording tip in the posterior thoracic oesophagus was first exposed to transmitted intraruminal pressure.

FIGURE 9.20

Intraluminal pressure changes of the thoracic oesophagus in response to injected rumen fluid during light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The arrow indicates an injection of rumen material (25.0 ml) into the mid-thoracic oesophagus. The injected rumen material induced a series of oesophageal peristaltic waves.

FIGURE 9.19

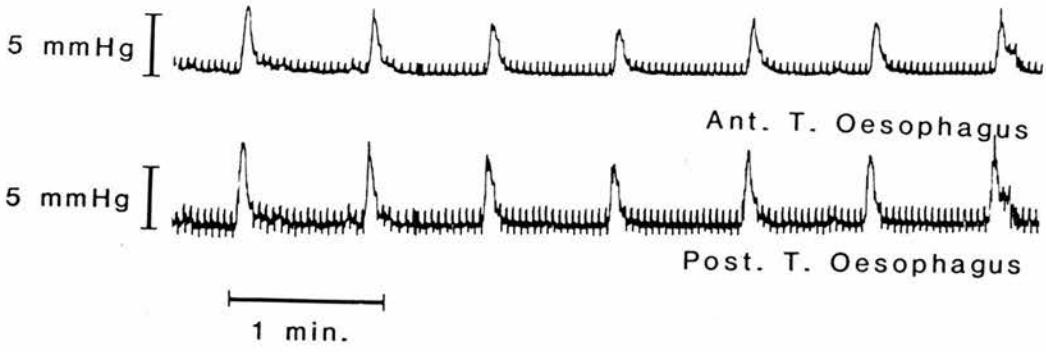
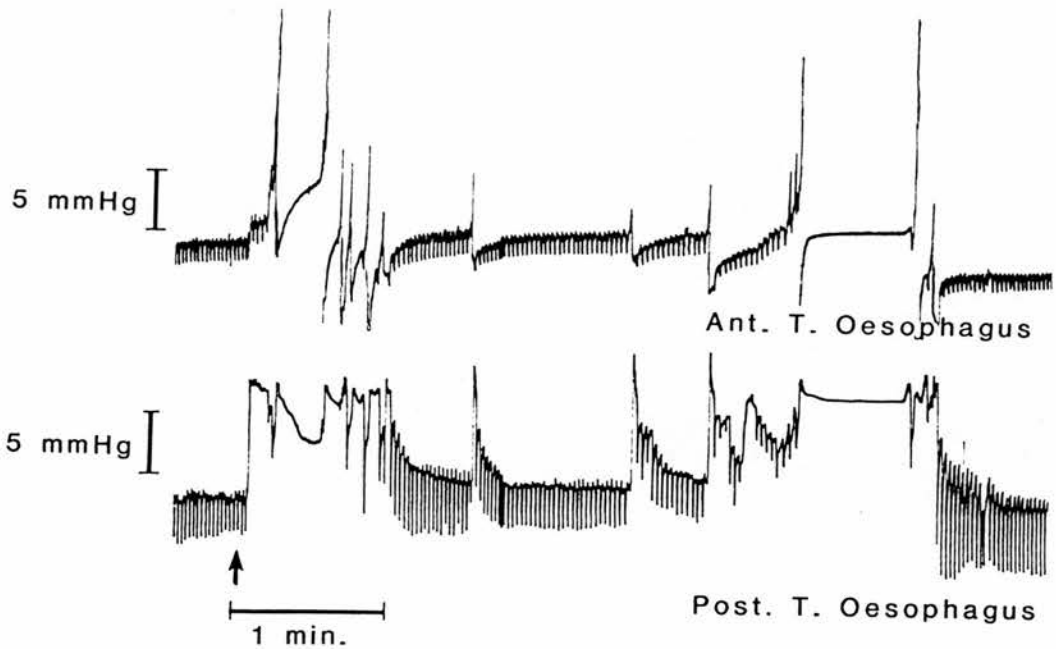


FIGURE 9.20



and 33.3% produced no change in light anaesthesia, while in deep anaesthesia, these figures were 57.1%, 0.0% and 42.9% respectively.

When a balloon was distended in the caudal oesophagus in light anaesthesia, the tone of the cervical oesophagus was increased to 22.2% of cases, decreased in 11.1% of cases and 66.7% underwent no noticeable change. These figures in deep anaesthesia were 28.6%, 28.6% and 42.9% respectively. The tone of the anterior thoracic oesophagus to such balloon distension was also influenced; in light anaesthesia, the tone was increased in 50% of cases, decreased in 16.7% of cases and no change in 42.9% of cases. In deep anaesthesia, these values were 78.6%, 14.3% and 7.1% respectively.

The response of anterior thoracic oesophagus to balloon distension in the caudal thoracic oesophagus was not consistent (Figure 9.21A). However, the pressure waves in the anterior thoracic oesophagus were consistently inhibited by balloon distension in the caudal oesophagus. The waves reappeared as soon as the balloon was deflated (Figure 9.21B). Prior to oesophageal contraction, the pressures were gradually increased. Following the wave of contraction, the tone of the oesophagus fell below the resting levels which again were gradually increased until another contraction occurred.

To summarise the results of balloon distension study, the tone of the thoracic oesophagus was predominantly increased due to balloon distension in the cervical oesophagus. The response of the cervical and anterior thoracic oesophagus due to balloon distension in the caudal oesophagus, however, was inconsistent.

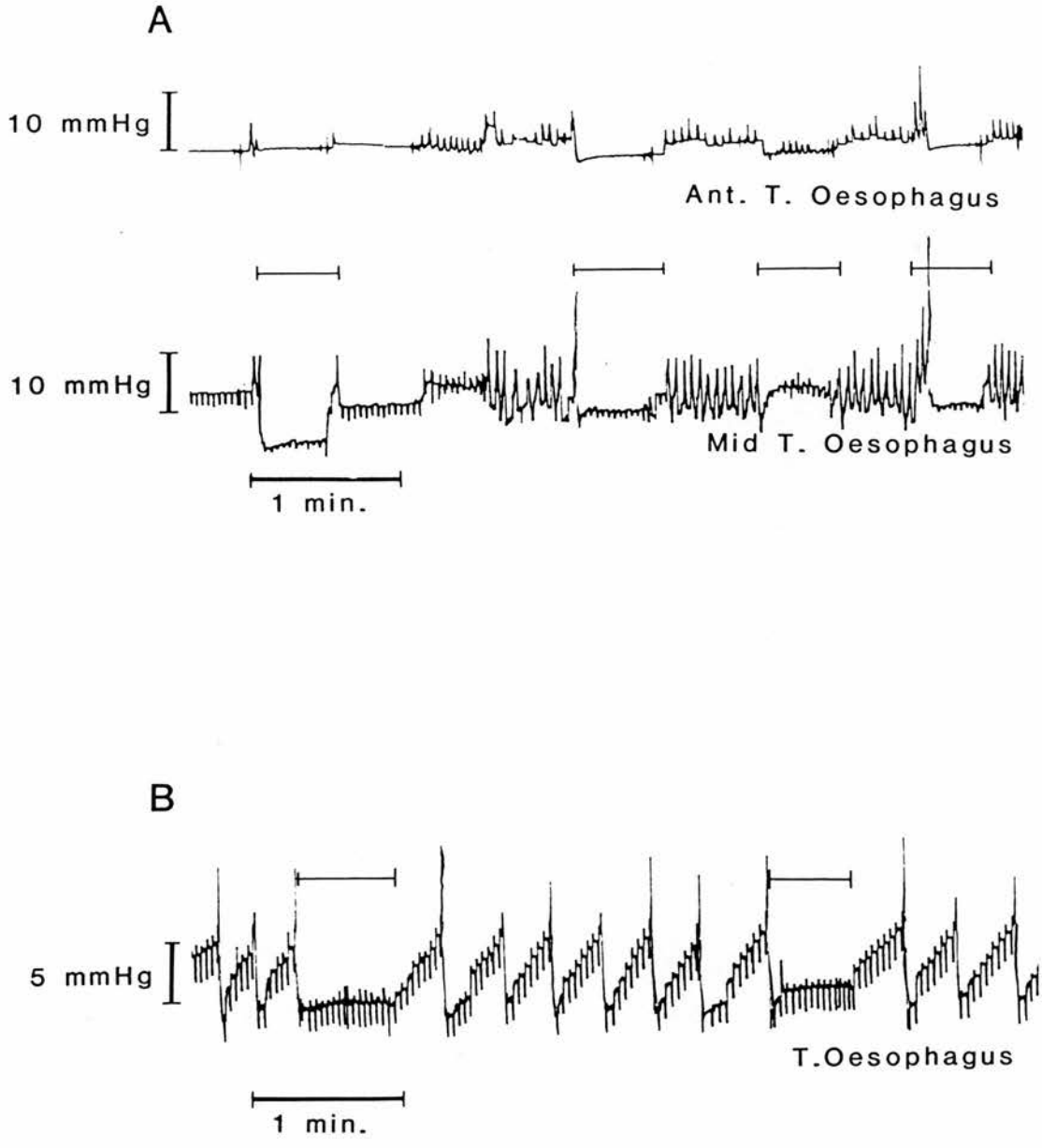
The response to balloon distension was difficult to evaluate due to marked variations in responses in individual sheep. For example sheep 645B was far less responsive than the other sheep.

FIGURE 9.21

A. Intraluminal pressure changes of the thoracic oesophagus in response to balloon distension in light anaesthesia (OTC). Upper tracing - anterior thoracic oesophagus (near thoracic inlet); lower tracing - mid-thoracic oesophagus (20.0 cm distal to thoracic inlet). The horizontal bars between the tracings indicate the duration of balloon distension (4.0 cm long balloon with 25.0 cc air) in the caudal oesophagus (within 10.0 cm of LOS). Note the inconsistent change in resting baseline pressure due to balloon distension.

B. Intraluminal pressure changes of the thoracic oesophagus in response to balloon distension in light anaesthesia. The top horizontal bars indicate the duration of balloon distension (4.0 cm long balloon with 25.0 cc air) in the caudal oesophagus (within 10.0 cm of LOS). The pressure waves present in the oesophagus were completely inhibited due to balloon distension.

FIGURE 9.21


A
10 mmHg
Ant. T. Oesophagus
10 mmHg
Mid T. Oesophagus
1 min.
B
5 mmHg
T.Oesophagus
1 min.

Discussion

The intraluminal manometry of rumen and oesophagus during anaesthesia provides information of their involvement in different physiological phenomena particularly associated with reflux and swallowing. The depth of anaesthesia affects the motility of reticulorumen and oesophagus. This is mainly due to the direct action of anaesthetic (halothane) on motor centres located in the medulla (Bell, 1961).

The intraluminal pressure changes in the oesophagus are due to direct contraction by the oesophageal muscle on the pressure sensitive catheter tip since the organ remains flaccid at rest. Therefore, the intra-oesophageal pressure reflects the motor activities of the organ due to tension developed in the circular muscle. The intra-oesophageal pressure closely reflects intrapleural pressure (Botha, 1959; Goyal and Cobb, 1981) although intra-oesophageal pressures are slightly higher than intrapleural pressures at the same site. The observations reported here confirm that the resting pressure in the body of the oesophagus was uniformly less than that in the rumen in the anaesthetized sheep.

The oesophagus is an organ which always remains empty at rest but is actively involved in different physiological events such as swallowing, regurgitation and eructation. During anaesthesia most of the reflexes for the above events are suppressed or abolished. The presence of pressure waves in the thoracic oesophagus appears to be due to excitation of the sensory receptors present in the oesophageal wall by the rumen contents since the saliva is drained off under such conditions. The occurrence of single oesophageal pressure waves possibly indicates an entry of a small amount of rumen contents

into the thoracic oesophagus while the group of pressure waves might indicate a moderate or larger reflux into the lower oesophagus.

The negative pressure in the oesophagus is probably maintained by the presence of pressure barriers at both ends, pharyngo-oesophageal sphincter and lower oesophageal sphincter (LOS). Swallowing, regurgitation and eructation are the events that involve the motility of the oesophagus. During anaesthesia, however, swallowing and eructation reflexes were suppressed unless the level of anaesthesia became very light.

The activities of the oesophagus and rumen depended on the level of central depression by halothane and may be associated with some reflux. The reflux of rumen contents into the thoracic oesophagus may cause mechanical stimulation which provides afferent inputs for oesophageal motor activities in light anaesthesia. The intensity of oesophageal pressure waves probably depends on the degree of mechanical stimulation and the level of anaesthesia. There are three possible mechanisms by which the oesophageal pressure waves can be initiated. These are (i) serial discharge of motor impulses from the medullary centre (Ingelfinger, 1958). This is probably suppressed in anaesthetized sheep, although this possibility has not been tested; (ii) as an integrated part of regurgitation, eructation and swallowing (Stevens and Sellers, 1968; Ali and Singleton, 1974; Weisbrodt, 1974). These reflexes during anaesthesia, however, are greatly suppressed by the anaesthetics (Meltzer, 1899; Botha, 1959). Moreover, no oesophageal antiperistalsis has been recorded associated with either "regurgitation" or "eructation" in these studies; (iii) the oesophageal pressure waves may be initiated by the presence of refluxed rumen material in the

thoracic oesophagus. This appears to be the most likely mechanism involved in initiating oesophageal pressure waves. The oesophageal pressure waves during anaesthesia have always been found to be peristaltic in these studies. This direction is probably physiological and ensures that the refluxed materials are swept back to the rumen. These peristaltic contractions in the lower oesophagus are probably stimulated by the refluxed rumen contents. The lower part of the oesophagus in sheep contains vagal mechanoreceptors (Falempin, Mei and Rousseau, 1978) which may be activated by mechanical distension caused by the refluxed material. The occurrence of thoracic oesophageal peristaltic waves were predominantly found in the distal part of the thoracic oesophagus which suggests that the reflex centre for the thoracic oesophagus is more irritable and offers greater resistance to the effects of anaesthesia. It is reported by Goyal and Cobb (1981) that food left behind in the oesophagus after primary peristalsis is cleared by secondary peristalsis. Also, when refluxed gastric contents appear in the distal oesophagus it is cleared by secondary peristalsis. This class of oesophageal pressure waves has been regarded as an indication of the occurrence of reflux in man (Creamer, 1955; Ingelfinger, 1958). It is probable that a similar mechanism exists in anaesthetized sheep.

The two most frequently used parameters for quantitative analysis of gastro-oesophageal motility records are the length or duration and the amplitude of the pressure waves. The amplitude of these waves can easily and accurately be determined; but the measurement of the duration of the waves often presents difficulties. In some cases, the waves occur slowly and in others very rapidly and do not return to

the baseline before the next one which renders inaccuracy in determining the actual measurement.

The mean amplitude of peristaltic contraction of the thoracic oesophagus not associated with reflux was greater than that associated with reflux. As oesophageal peristaltic waves might indicate the occurrence of reflux in a light plane of anaesthesia, the greater amplitude of thoracic oesophageal pressure waves may be necessary to clear the refluxed rumen material. When these pressure waves are weaker, the rumen contents may overcome the barrier and be carried to the oro-pharynx in head down position.

Usually, the amplitude of peristaltic contractions in the posterior thoracic oesophagus is greater than those in the anterior part during light anaesthesia. These findings are similar to those reported in man (Botha, 1959; Humphries and Castell, 1977).

The amplitude of contraction of oesophagus containing exclusively smooth muscle (e.g. opossum, man etc.) appears to be greater. In man, the mean amplitude of oesophageal contraction has been reported to be 85 mm Hg (Mellow, 1983) while their duration was 3.65 seconds. The longer duration indicates a slower speed of peristalsis. In the present investigation, the mean amplitude and duration of peristaltic waves in the posterior thoracic oesophagus in the light plane of anaesthesia were 15.2 mm Hg and 1.4 seconds respectively. The shorter duration of oesophageal peristalsis in ruminants indicates a faster velocity and it is probably related to striated muscle in these species. On the other hand, a relatively weaker amplitude of oesophageal pressure wave indicates that contractile strength of circular muscle (striated) in sheep is limited under anaesthesia.

In deep anaesthesia, no contraction was usually noticed either in the oesophageal or ruminal tracing. However, the baseline pressure of thoracic oesophagus occasionally increased for a variable length of time. The increased tone might function to protect the oesophagus from reflux.

The thoracic oesophageal pressure waves were usually independent of rumen contractions; but in a few cases, the oesophageal contractions preceded the ruminal ones with comparatively greater amplitudes (Figure 9.8). This response might result from the displacement of the recording tip towards the LOS.

The cervical oesophagus in anaesthetized sheep appeared to be less active when compared with the thoracic oesophagus suggesting that the cervical oesophagus is relatively less sensitive in such conditions. The reason may be that the refluxed rumen material which appears to be the major stimulus for initiating secondary peristalsis in the thoracic oesophagus only occasionally reaches the cervical level. Alternatively, the excitatory receptors for this reflex peristalsis are predominantly present in the thoracic oesophagus.

From these results, it is concluded that light anaesthesia is relatively safer than deep anaesthesia from the standpoint of reflux; because in light anaesthesia, if there is any reflux in the lower oesophagus, it is likely to be driven back into the rumen by secondary oesophageal peristalsis which still remains active at this depth of anaesthesia. In a deep plane of anaesthesia, however, since the oesophageal reflexes are greatly suppressed, any rumen material appearing in the lower oesophagus usually does not provoke any defensive peristalsis and eventually the refluxed rumen contents flow out through

the pharynx if the front of the animal is tilted down. Passage of rumen contents into the thoracic oesophagus does not seem to be harmful unless these materials proceed further out into the oro-pharynx. Unlike monogastric animals, the rumen contents do not cause irritation to the oesophagus since the pH of the rumen contents is usually maintained near the neutral.

In conscious sheep, a healthy rumen exhibits three contractions in two minutes (Leek, 1983). However, when the animal is maintained under halothane anaesthesia, both the rate and amplitude of contractions are greatly suppressed and eventually abolished in deep anaesthesia. The progressive suppression of rumen contractions due to increasing concentration of halothane suggests that the irritability of the reticuloruminal motor centre in the medulla is inversely related to the concentration of halothane in the blood circulation. The absence of these contractions in anaesthetized sheep has also been reported elsewhere (Duncan, 1951; Phillipson, 1970). This eliminates the possibility of any active reticuloruminal and medullary involvement at reflux during deep anaesthesia. Hence the mechanism of reflux during anaesthesia is different from that of regurgitation in conscious animals where the contraction of reticulorumen is an essential component co-ordinated by medullary centres.

The rumen contractions were frequently present during light anaesthesia although in some cases these contractions were very poor or absent. This may be due to individual variation or the physical condition of the rumen. It has been observed clearly that the motility of rumen and oesophagus can occur independently which suggests that the rumen and oesophagus have discrete controlling mechanisms. The

co-ordinated activities of these organs, however, are observed in the conscious state in regurgitation, eructation etc.

In starved animals, the rumen contractions were weak or absent which presumably was due to the significant reduction in the rate of intraruminal gas production (Chapter 7). This reduced amount of gas is perhaps not sufficient to stimulate the tension receptors of the reticulorumen (Iggo, 1954; Falempin et al., 1978). This may also be due to the physical state of the ingesta. Benzie and Phillipson (1957), however, reported that movements of the reticulorumen can occur in young suckling animals even though these organs contain no solid food.

From the intraluminal pressure studies of the rumen and oesophagus it has been observed that two completely different pressure profiles are present in the above organs although there is no anatomical partition between them. These manifestations obviously put forward the possibility of a unique device between the reticulorumen and oesophagus that remains impatent and thus enables the maintenance of discrete pressure profiles on either side. When this device becomes patent, the positive intraruminal pressure is transmitted to the negative intra-oesophageal pressure. Complete sphincteric patency was observed in one anaesthetic during the course of the investigation where the pressure profiles of both rumen and oesophagus became identical and there was profuse reflux (Figure 9.19).

No oesophageal antiperistalsis was observed during gaseous reflux in anaesthetized sheep in contrast to the conscious state, therefore eructation did not occur during anaesthesia. In a few sheep, the rumen gases were found to be expelled through the mouth with audible sound

and characteristic odour. The pressure changes in the oesophagus during this event, however, were peristaltic. During expulsion of gas, some rumen contents might enter the oesophagus which in turn may stimulate the vagal mechanoreceptors for a reflex oesophageal peristalsis.

The oesophageal response to balloon distension in both light and deep planes of anaesthesia was not consistent. It is reported in conscious sheep that distension of a balloon in the distal one-third of the oesophagus initiates secondary peristalsis distal to the point of distension (Winship et al., 1964). This was, however, not observed in anaesthetized sheep in the present experimental conditions. The reasons are not clearly understood. In monogastric animals and man, balloon distension in the oesophagus also provokes peristaltic contraction (Creamer and Schlegel, 1957; Code et al., 1958).

Conclusions

(i) Oesophageal and reticuloruminal motility are present in light anaesthesia. These activities in deep anaesthesia, however, are mostly inhibited. The investigation suggests that oesophagus and reticulo-rumen have discrete controlling mechanisms.

(ii) Oesophageal activities during anaesthesia are also peristaltic. These peristaltic pressure waves might indicate the presence of refluxed material.

(iii) Reflux during anaesthesia is passive and it was not associated with any oesophageal antiperistalsis.

(iv) The swallowing reflex is present in light anaesthesia, but is totally inhibited in the deep plane of anaesthesia.

(v) Light anaesthesia may be less hazardous than deep anaesthesia; because in light anaesthesia, the protective reflex (oesophageal peristalsis) is present.

(vi) There is a functional barrier at the gastro-oesophageal junction in anaesthetized sheep.

(vii) Oesophageal response to balloon distension is variable.

CHAPTER TEN

A MANOMETRIC STUDY OF THE ACTIVITIES OF LOWER OESOPHAGEAL SPHINCTER

Introduction

It has been demonstrated in an insufflation study (Chapter 8) that the amount of intraruminal pressure required to produce gastro-oesophageal reflux is 4-5 times (32-42 mmHg), the intraruminal pressures found during anaesthesia lasting up to 2-3 hours. Also, it was demonstrated in Chapter 7 that two completely different pressure profiles exist on either side of the gastro-oesophageal junction. These findings suggest that some mechanism must exist at the junction which controls the movement of rumen materials to and from the oesophagus. It is generally accepted that in monogastric animals (e.g. dog, opossum, man) the barrier at this junction is maintained by a high pressure zone (HPZ) which has been called the lower oesophageal sphincter (LOS). Studies on LOS in ruminants, however, are limited. Winship et al. (1964) were unable to demonstrate a HPZ or LOS at the gastro-oesophageal junction in the conscious sheep. Dougherty and Meredith (1955) demonstrated radiographically an oesophageal constriction cranial to the diaphragm which they called the prediaphragmatic sphincter. They did not, however, perform any manometry and so it is not known whether this sphincter maintains any high pressure zone. There appears to have been no reported studies of the role of the lower oesophageal sphincter in the control of the movement of ingesta in the ruminant in either conscious or anaesthetized animals. Experiments were therefore designed and performed to demonstrate the presence or absence of a high pressure zone at the gastro-oesophageal junction and to characterise the activities of this area in sheep during anaesthesia.

Materials and Methods

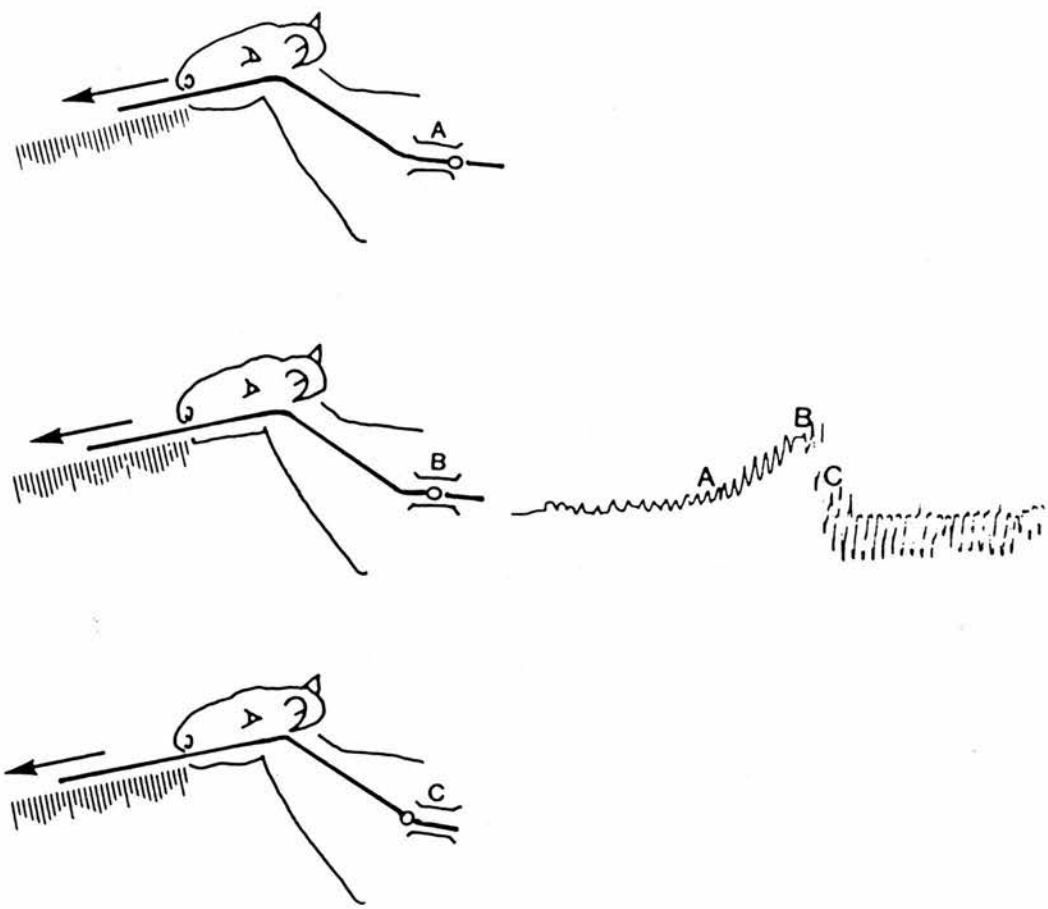
Eleven sheep were used in this investigation. Induction of anaesthesia was performed with a mixture of halothane (4%), nitrous oxide and oxygen (50:50) using a face mask and a Magill system. After induction, anaesthesia was maintained as described previously (Chapter 4). The animals used in this study were not starved and were positioned in right lateral recumbancy.

Detection and Length of LOS

The existence and length of LOS were investigated using a balloon tip catheter (1.0 cm long balloon) by employing catheter assembly 1 (Chapter 4). The catheter assembly was introduced through the pharynx and oesophagus so that the balloon was in the rumen. The balloon was then inflated with 0.2-0.3 cc air using a 2.0 ml plastic syringe. A ruler was held against the incisor teeth in close proximity to the catheter assembly (Figure 10.1). The catheter assembly was then gradually withdrawn and as the balloon passed through the gastro-oesophageal junction, any change in baseline pressure was monitored. The existence of the LOS was detected by the appearance of a high pressure zone and the length of the sphincter was delineated by that portion of the withdrawal tracing that indicated pressures greater than those obtained in the rumen.

After the recognition of a high pressure zone, the functional behaviour of the LOS was investigated by continuous monitoring of LOS pressure using a water filled open tip catheter and "stationary technique". In this technique, the recording tip was continuously maintained in the LOS and any displacement of the catheter was restored

FIGURE 10.1
Technique for measuring the lower oesophageal sphincter length using continuous slow pull through method.



The balloon catheter was first pushed down to the rumen and then it was withdrawn. At A, the balloon was entering the LOS, at B, it was in the middle of the LOS and at C, the balloon was just in the thoracic oesophagus. The length of LOS was measured from the distance A to C from the ruler placed against the incisor teeth.

by moving the catheter up or down. The cranial displacement of the catheter was recognised by the appearance of negative intrathoracic pressure and was characterised by the downward deflection of the respiratory pressure changes. The caudal displacement of the catheter was identified by the presence of characteristic ruminal pressure changes with a stable resting baseline. The LOS pressure changes were characterised by its rapidly fluctuating baseline pressure. The patency of the catheter was occasionally impaired (evident from the absence of respiratory pressure changes and reduction of baseline pressure) and was restored by flushing the catheter with a few millilitres of water. Once the positioning of the recording tip in the LOS was achieved, the pressures were monitored over a period of 60 minutes.

The LOS pressure was recorded using both balloon tip and open tip catheters employing the catheter assembly 1 and 2 as described in the general methods (Chapter 4).

Influence of Depth of Anaesthesia on LOS

The influence of the depth of anaesthesia on LOS pressure was studied using two planes of anaesthesia, light (concentration of halothane in the expired gases between 0.5-1.0%) and deep (concentration of halothane in the expired gases between 1.5-2.0%) were maintained for two 30-minute periods in a single anaesthetic session. The selection of the plane of anaesthesia for the first 30 minutes was random.

Effect of Intraruminal Pressure on LOS

The response of LOS to an increased intraruminal pressure was

measured in a series of experiments. The intraruminal pressure was manipulated in a controlled manner by applying external pressure on the abdomen. The technique used for this study included a piece of canvas which was wrapped around the abdomen of the anaesthetized sheep. Both ends of the canvas were tied with ropes across the operating table and the ropes were twisted with a piece of wood to compress the abdomen for elevating the intra-abdominal and intra-ruminal pressure.

Effect of Oeosphageal Distension on LOS

The response of LOS to intraluminal distension of a balloon was studied by using a separate catheter assembly (Chapter 4). The catheters were arranged so that a balloon (4.0 cm long) could be distended (with 25 cc air) in either the thoracic or cervical oesophagus while the responses of LOS were recorded. The data obtained in these studies were analysed by using the Student's t-test, where appropriate.

Results

Detection and length of LOS: Withdrawal of the catheter assembly across the gastro-oesophageal junction recorded a zone of higher pressure than that of the rumen immediately before the low pressure of the oesophagus. A high pressure zone at the gastro-oesophageal junction was always recorded using a balloon tip catheter and the zone was well represented in this technique (Figure 10.2). A high pressure zone was not, however, consistently recorded with an open tip catheter. The withdrawal tracings with the open tip catheter frequently failed to identify the high pressure at the gastro-oesophageal junction.

However, by repeating the manoeuvre it was always possible to detect this zone in every anaesthetic although the zone of elevated pressure was less obvious using this technique (Figure 10.3). As the recording catheter was withdrawn through the gastro-oesophageal junction, there was usually a gradual rise in the baseline pressure with increased respiratory excursions. As the catheter tip passed the LOS into the thoracic oesophagus, the high pressure dropped sharply to the negative intrathoracic pressure with respect to atmosphere.

The reverse manoeuvre i.e. the advancement of a recording tip from the oesophagus into the LOS also showed the high pressure zone (Figure 10.4). As the recording catheter entered the LOS, the swing of the negative intrathoracic pressure was changed to a positive intra-sphincteric pressure.

In two anaesthetics, slow gastro-oesophageal withdrawal tracings showed consistently a two-stage high pressure zone (Figure 10.5). The withdrawal tracings in these cases recorded a zone of high pressure and then the pressure dropped to near the atmospheric pressure and recorded another similar zone of high pressure as the catheter was further withdrawn and finally the pressure dropped to negative intra-thoracic pressure.

This withdrawal manometry confirmed the existence of a high pressure zone, possibly associated with lower oesophageal sphincter in the anaesthetized sheep. The length of this sphincter was measured and the results are presented in Table 10.1. The length of zone of high pressure was 2.9 ± 0.3 (SD) cm.

Table 10.1 The length of lower oesophageal sphincter during halothane anaesthesia using balloon tip catheter and pull through method

Sheep No.	Lower Oesophageal Sphincter Length (cm)					
	Single values					Mean values
	1	2	3	4	5	
567E	3.0	2.9	2.7	2.0	2.5	2.62
566E	2.5	3.0	3.5	3.8	4.0	3.36
518E	2.5	2.6	1.8	3.2	2.4	2.50
Z650	2.6	1.9	2.9	2.2	2.7	2.46
127B	2.9	3.0	3.2	2.8	3.1	3.0
621E	2.5	3.0	2.5	3.0	4.0	3.0
594E	5.5	3.0	2.5	3.0	1.5	3.1
645B	2.8	4.0	3.5	2.5	3.0	3.16
Total mean						2.9
S.D.						0.33

FIGURE 10.2

The change in pressure associated with LOS as detected in manometry using air filled balloon tip catheter and pull through technique. The length of balloon was 1.0 cm and inflated with 0.2-0.3 cc room air. As the recording balloon was withdrawn through the LOS, a zone of high pressure (HPZ) was recorded.

FIGURE 10.3

LOS as appeared in manometry using water filled open tip catheter and pull through technique. As the recording tip passed through the LOS, respiratory pressure fluctuations became prominent. The high pressure zone (HPZ) in this technique was not obvious.

FIGURE 10.4

LOS as appeared in reverse manoeuvering of catheter assembly (balloon tip). As the recording balloon was introduced through the oesophagus into the LOS, the swing of the respiratory pressure changes was reversed (downward arrow). Again, as the recording tip was withdrawn from the LOS into the thoracic oesophagus, similar changes occurred (upward arrow). The point where the swing of the respiratory pressure changes was reversed is called the point of respiratory pressure reversal.

FIGURE 10.5

Two-stage high pressure zone as recorded using balloon tip catheter and pull through technique. As the recording balloon was withdrawn through the LOS, a first zone of high pressure (HPZ-1) appeared which dropped almost to the baseline (intraruminal) pressure and again a second zone of high pressure (HPZ-2) was noticed.

FIGURE 10.2

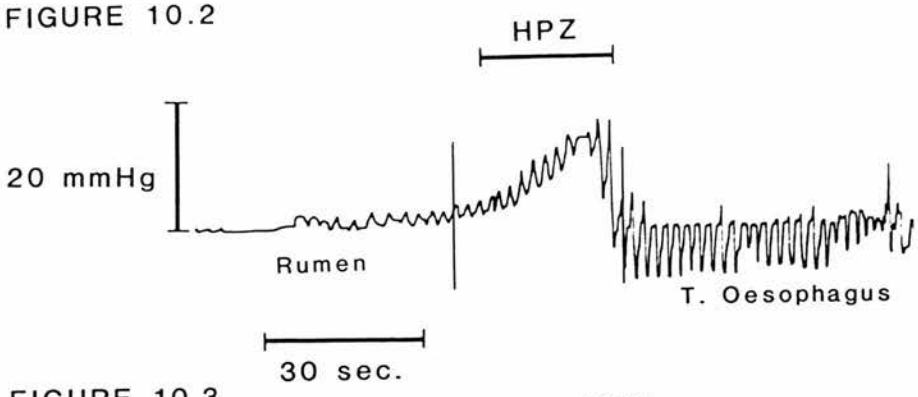


FIGURE 10.3

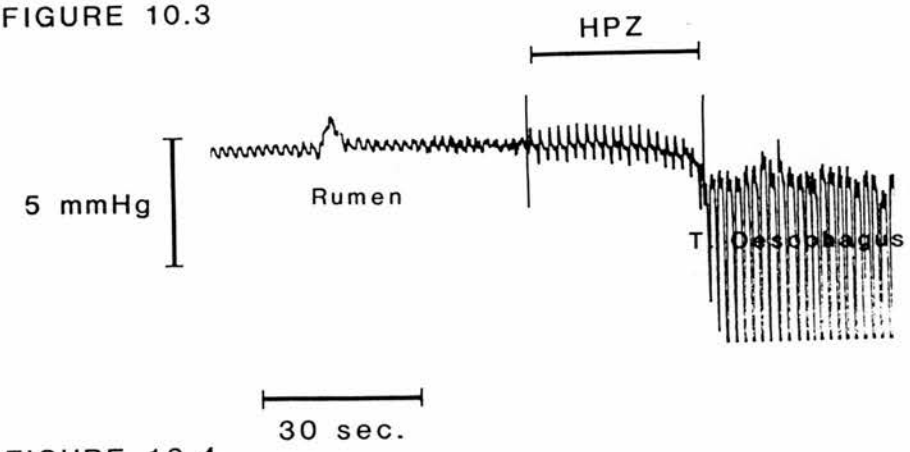


FIGURE 10.4

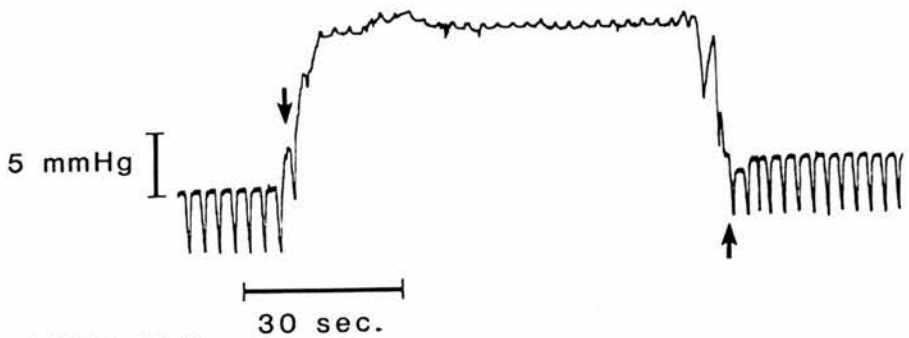
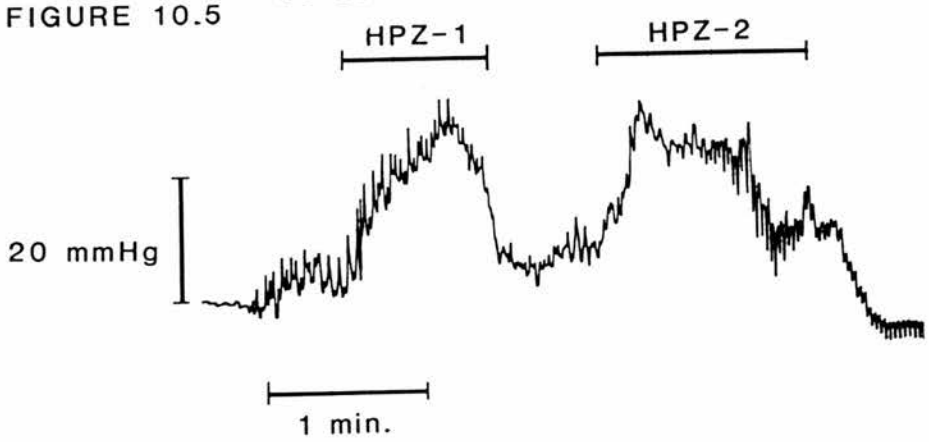


FIGURE 10.5



Resting LOS pressure: The resting intraluminal pressure of LOS was recorded using both balloon tip and open tip catheters. The results of these studies are presented in Tables 10.2 and 10.3 respectively. The resting LOS pressure using balloon tip catheter was 25.6 ± 2.6 (SD) mmHg and that using open tip catheter was 14.3 ± 2.5 (SD) mmHg. On statistical analysis, this difference was highly significant ($P < 0.001$). The rumen pressures in these studies were also measured using open tip catheters and these values were 10.01 ± 1.9 (SD) mmHg and 13.6 ± 2.4 (SD) mmHg respectively. The LOS-rumen pressure gradient using balloon catheter (for LOS only) was 15.6 ± 3.3 (SD) mmHg and that using open tip catheter was 0.70 ± 1.02 (SD) mmHg. This difference was also highly significant ($P < 0.001$). The resting pressure of LOS was more variable than that of either oesophagus or rumen.

Effects of depth of anaesthesia on resting LOS tone: This was studied using open tip catheters. The results are displayed in Table 10.4. The resting intraluminal pressure of LOS in light anaesthesia was 11.5 ± 3.09 (SD) mmHg and that in deep anaesthesia was 10.9 ± 2.6 (SD) mmHg. The difference was not statistically significant ($P > 0.05$).

Patterns of LOS pressure changes: The characteristic patterns of LOS pressure changes associated with ruminal and oesophageal activities were studied. Simultaneous recording of pressures from the LOS and rumen showed a unique relationship. In about 80% of cases, the rumen contractions were preceded by LOS contractions. About a second prior to rumen contraction, an increase in LOS pressure usually

Table 10.2 Resting pressure of rumen and LOS during halothane anaesthesia using balloon tip catheter

Sheep No.	Resting pressure Rumen (mmHg) n = 4	Resting pressure LOS (mmHg) n = 4	LOS - Rumen gradient (mmHg)
567E	9.25	25.87	16.62
566E	10.25	29.50	19.25
518E	14.0	25.87	11.87
Z650	9.5	24.75	15.25
127B	7.62	28.75	21.13
621E	9.5	22.0	12.5
594E	9.0	22.5	13.5
645B	11.0	25.5	14.5
Mean	10.01	25.59	15.57
S.D.	1.88	2.63	3.25

Table 10.3 Resting pressure of rumen and LOS during halothane anaesthesia using open tip catheter.

Sheep No.	Resting pressure Rumen (mmHg) n = 10	Resting pressure LOS (mmHg) n = 10	LOS - Rumen gradient (mmHg)
566E	13.35	15.05	1.7
552E	11.35	13.5	2.15
Z650	17.55	18.35	0.8
567E	12.4	12.45	0.05
Z981	15.2	15.0	-0.20
645B	11.5	11.25	-0.25
Mean	13.55	14.26	0.70
S.D.	2.41	2.48	1.02

Table 10.4 Lower oesophageal sphincter pressure during light and deep planes of halothane anaesthesia using open tip catheter

Anaesthetic No.	Lower Oseophageal Sphincter Pressure (mmHg)									
	Light plane readings				Mean values	Deep plane readings				Mean values
	1	2	3	4		1	2	3	4	
1	9.0	9.0	11.0	7.0	9.0	7.0	8.5	9.0	12.0	9.12
2	6.0	5.0	17.5	12.0	10.12	12.0	12.0	9.0	7.5	10.12
3	10.0	10.0	10.0	10.0	10.0	10.0	7.5	9.0	10.0	9.12
4	13.5	15.5	15.5	15.0	14.87	15.0	12.5	15.0	14.5	14.25
5	3.5	4.5	7.5	6.0	5.37	6.0	7.0	6.0	6.5	6.37
6	15.0	15.0	15.0	12.5	14.37	12.5	7.5	12.0	12.0	11.0
7	11.5	11.0	10.0	9.0	10.37	9.0	11.0	8.0	8.5	9.12
8	15.0	15.0	13.5	15.0	14.62	15.0	15.0	13.0	13.5	14.12
9	15.0	12.5	12.5	11.0	12.75	11.0	12.0	15.0	15.0	13.2
10	13.0	13.0	14.0	16.0	14.0	12.5	14.0	12.5	13.0	13.0
Mean					11.54					10.94
S.D.					3.09					2.62

P > 0.05

N.B. The lower oesophageal sphincter pressure was monitored over a period of 60 minutes during halothane anaesthesia (light followed by deep and vice-versa). The four readings in each plane were taken at 0, 10, 20 and 30 minutes of anaesthesia.

occurred with amplitude 4-5 times greater than those of ruminal pressure waves (Figure 10.6). This pattern of LOS activity, however, was not uniform and varied between different sheep and also during the same anaesthetic. Sometimes the LOS contractions were weak or absent in association with rumen contractions (Figure 10.7). Although the LOS contractions usually preceded the rumen contractions, in about 10% of cases the LOS relaxed prior to rumen contraction (Figure 10.8). Again in about 10% of cases, LOS relaxation occurred after rumen contraction (Figure 10.9). No change in thoracic oesophageal pressure was detected during the rhythmic fluctuations of LOS pressure (Figure 10.10). However, when pressure waves were present in the oesophagus, the LOS showed an early relaxation which was followed by a contraction (Figure 10.11).

As anaesthesia deepened, the amplitudes of LOS and ruminal pressure waves were progressively diminished, but unlike rumen the LOS activities were not completely abolished (Figure 10.12). When ruminal pressure waves were completely abolished, the LOS still showed rhythmic fluctuations in its resting pressure (Figure 10.13). During deep anaesthesia, the LOS occasionally showed relatively longer phases of relaxation (Figure 10.14).

The intraluminal pressure changes of LOS associated with reflux were monitored in light anaesthesia. The tone of LOS was reduced during reflux usually below the intraruminal pressure (Figure 10.15). The diminished sphincteric pressure associated with reflux in light anaesthesia was frequently recovered following an oesophageal peristaltic wave (Figure 10.16). A relaxation of LOS was also associated with reflux in deep anaesthesia (Figure 10.17). This reflux-associated LOS relaxation, however, was not consistently demonstrated.

FIGURE 10.6

Activity of LOS during rumen contraction in the light plane of anaesthesia using water filled open tip catheter (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Just before (about a second) rumen contraction (upward deflections of baseline pressures) the LOS contracted usually with a higher amplitude.

FIGURE 10.7

LOS and rumen pressure changes in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Note that the LOS did not contract consistently (the arrows indicate LOS contractions).

FIGURE 10.6

FIGURE 10.7

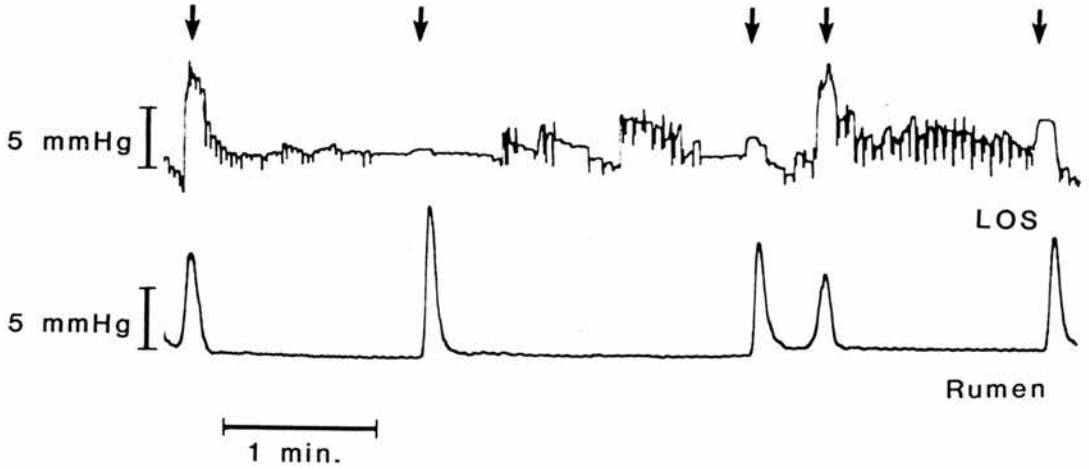


FIGURE 10.8

LOS and rumen pressure changes in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The LOS showed a variable degree of relaxation before rumen contraction (arrows).

FIGURE 10.9

LOS and rumen pressure changes in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Note the relaxation of LOS after rumen contraction. The arrows indicate the LOS relaxations.

FIGURE 10.8

FIGURE 10.9

FIGURE 10.10

Resting pressure of thoracic oesophagus and LOS in light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - LOS. The LOS tracing showed rhythmic fluctuation in pressure while the thoracic oesophagus was quiescent except for respiratory changes.

FIGURE 10.11

Thoracic oesophageal and LOS pressure changes in light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - LOS. The oesophageal pressure waves were associated with LOS relaxations which were followed by contraction.

FIGURE 10.10

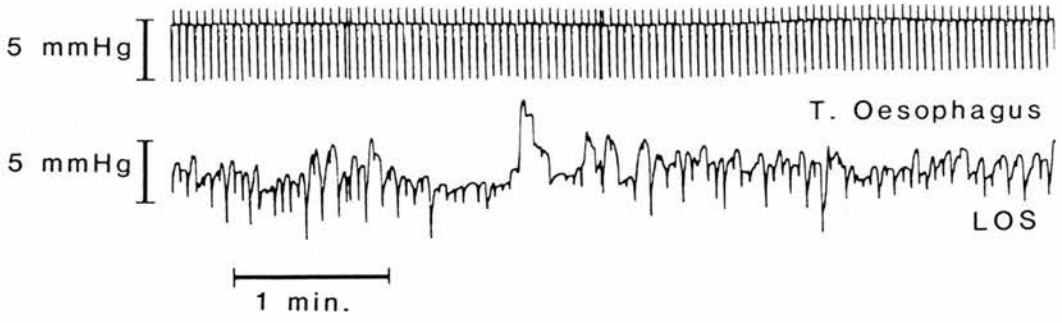


FIGURE 10.11

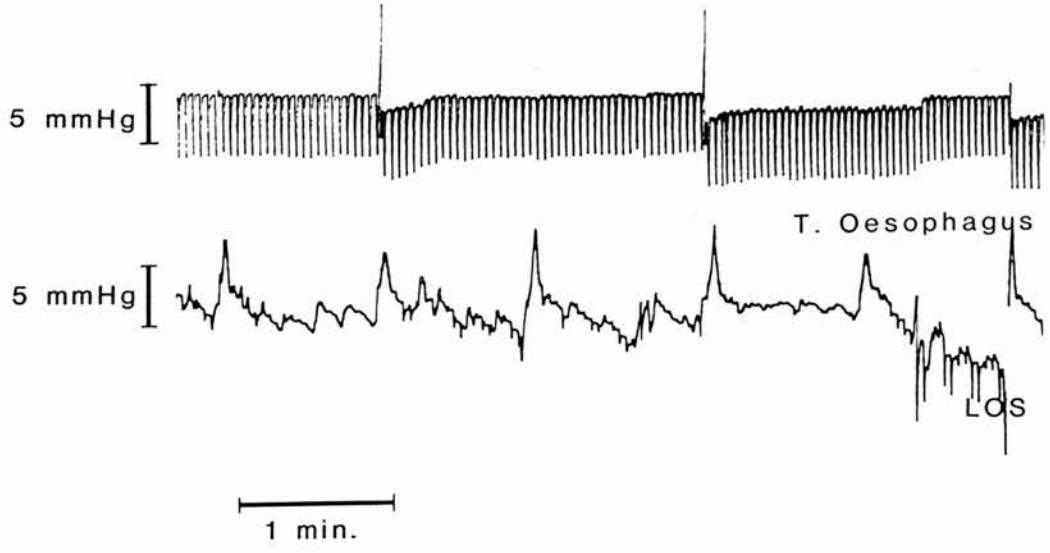


FIGURE 10.12

LOS and rumen pressure changes as anaesthesia deepened (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). With the increase in the depth of anaesthesia, the rumen contractions progressively disappeared. The LOS response, on the other hand, showed a little change in pressure.

FIGURE 10.13

LOS and rumen pressure changes in deep anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The rumen contractions were completely abolished whereas LOS showed frequent changes in pressure.

FIGURE 10.14

LOS and rumen pressure changes in deep anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Note the prolonged relaxation of LOS tone. These relaxations appeared to be associated with extremely weaker rumen activity (arrows).

FIGURE 10.12

FIGURE 10.13

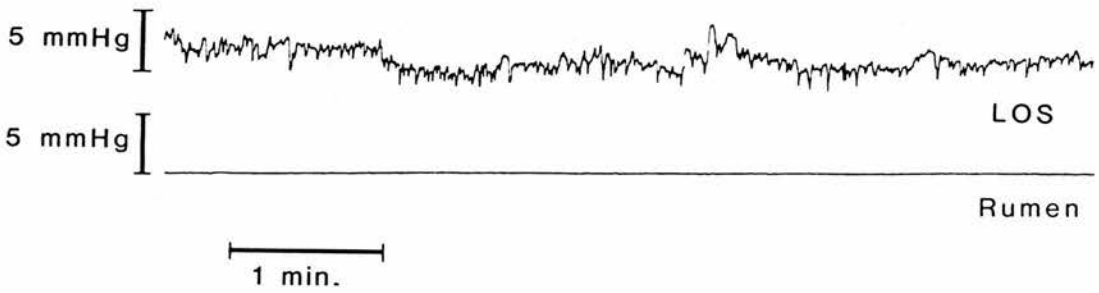


FIGURE 10.14

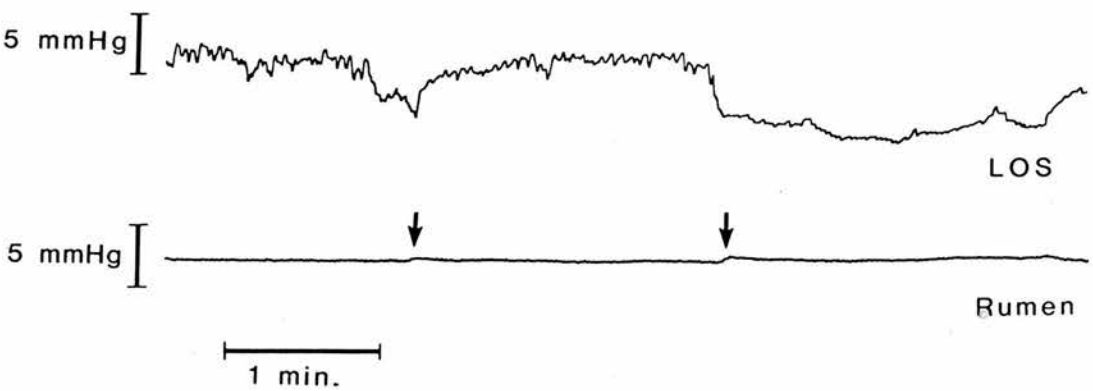


FIGURE 10.15

LOS and rumen pressure changes associated with reflux in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the onset (upward arrow) and termination (downward arrow) of reflux. With the onset of reflux, the LOS tone was reduced but was recovered after the termination of reflux. The LOS did not show any activity in response to rumen contractions. The rumen contractions, however, did not appear to be involved in such reflux.

FIGURE 10.16

Thoracic oesophageal and LOS pressure changes associated with reflux in light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - LOS. The arrows indicate the occurrence of mild and transient reflux which were coincided with smaller relaxations in LOS pressure. The relaxed LOS tone, however, was recovered following oesophageal peristaltic wave. The presence of interrupted respiratory pressure changes in these tracings was due to cessation of breathing.

FIGURE 10.17

LOS pressure changes associated with reflux in deep anaesthesia (OTC). The arrows indicate the occurrence of mild and transient reflux. The sphincteric relaxation was consistently present at the event of reflux.

FIGURE 10.15

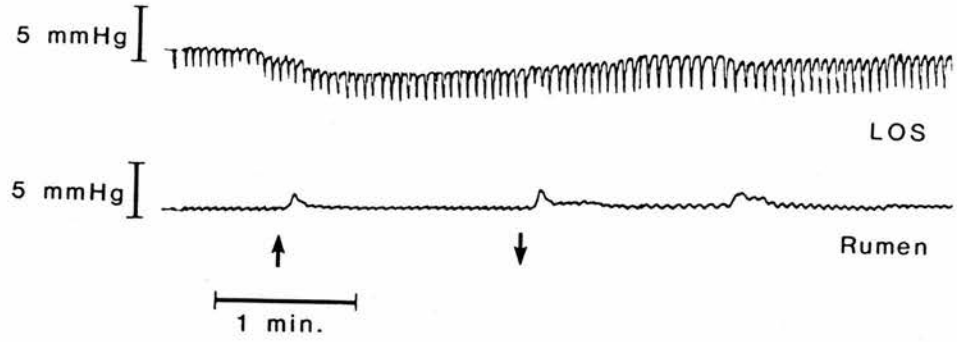


FIGURE 10.16

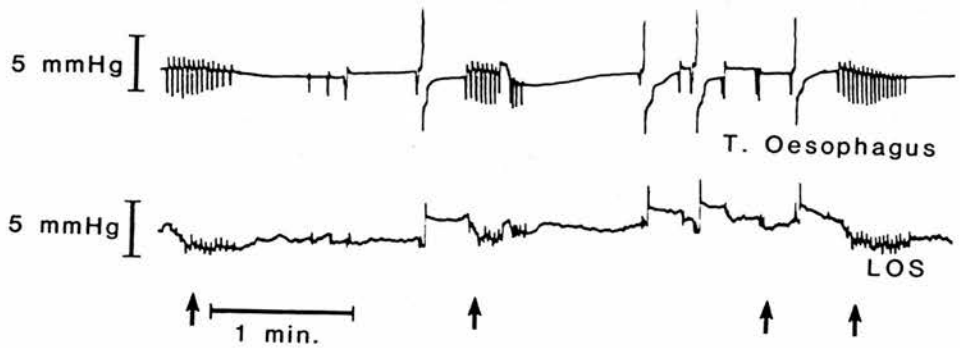
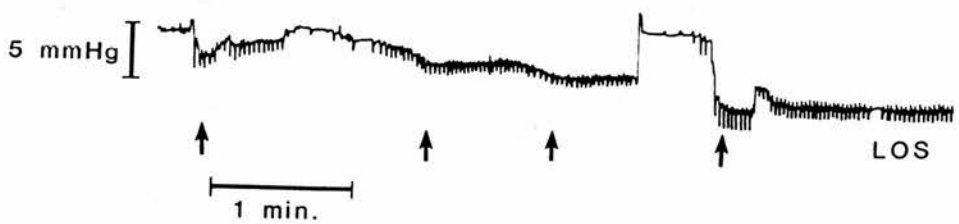


FIGURE 10.17



Effect of increasing rumen pressure: The response of LOS to an increase in intraruminal pressure was studied. The intraruminal pressure was increased by controlled steps of 5 and 10 mmHg to observe the response of LOS pressure in both light and deep planes of anaesthesia. The LOS pressure was increased at the same time as an increased intraruminal pressure. The results are presented in Table 10.5. An elevation of intraruminal pressure by 5.0 mmHg in light anaesthesia caused the LOS pressure to be increased by 5.4 ± 1.05 (SD) mmHg and that in deep anaesthesia by 5.8 ± 0.69 (SD) mmHg. The difference was not significant. Again, an elevation of intraruminal pressure by 10.0 mmHg in light anaesthesia caused the LOS pressure to be increased by 10.4 ± 0.68 (SD) mmHg and that in deep anaesthesia by 10.4 ± 0.94 (SD) mmHg. This difference was also not significant. The response of LOS to increased intraruminal pressure is shown in Figures 10.18 and 10.19. The oesophageal pressures during these manoeuvres showed an increase in baseline pressure (Figure 10.20).

Effect of oesophageal distension on LOS: The response of LOS to intraluminal balloon distension in the oesophagus was studied in both light and deep planes of anaesthesia. In light anaesthesia, distension of a balloon in the thoracic oesophagus caused relaxation of LOS in 75% of cases. When a rumen contraction occurred during balloon distension, the LOS still contracted in a similar characteristic fashion with the same degree of amplitude as when the LOS relaxation was not provoked (Figure 10.21). With similar distension of a balloon in deep anaesthesia, the sphincter tone was usually increased (Figure 10.22).

Table 10.5 Response of LOS to an increase of intraruminal pressure caused by abdominal compression

Sheep No.	Degree of abdominal compression above the resting pressure (mmHg) n = 5	Responded LOS Tone (mmHg) n = 5	
		Light plane	Deep plane
566E	5.0	7.3	5.5
	10.0	10.8	10.0
522E	5.0	5.5	5.7
	10.0	11.0	12.2
Z650	5.0	5.3	5.3
	10.0	10.2	10.0
567E	5.0	5.1	6.6
	10.0	10.8	10.6
Z981	5.0	5.0	5.0
	10.0	9.5	9.8
645B	5.0	4.1	6.7
	10.0	9.8	9.7
Mean	5.0	5.38	5.8
	10.0	10.4	10.38
S.D.		1.05	0.69
		0.68	0.94

FIGURE 10.18

The response of LOS to an increased intraruminal pressure in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The intraruminal pressure was increased by 5.0 mmHg indicated by arrows. The LOS also showed pressure waves associated with increased intraruminal pressure usually with larger amplitude.

FIGURE 10.19

The response of LOS to an increased intraruminal pressure in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The intraruminal pressure was increased by 10.0 mmHg indicated by arrows. The LOS showed a larger amplitude pressure change associated with increased intraruminal pressure.

FIGURE 10.20

The response of thoracic oesophagus to an increased intraruminal pressure (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The intraruminal pressure was increased by 5.0 mmHg indicated by arrows. The thoracic oesophagus showed an increase in tone associated with increased intraruminal pressure.

FIGURE 10.18

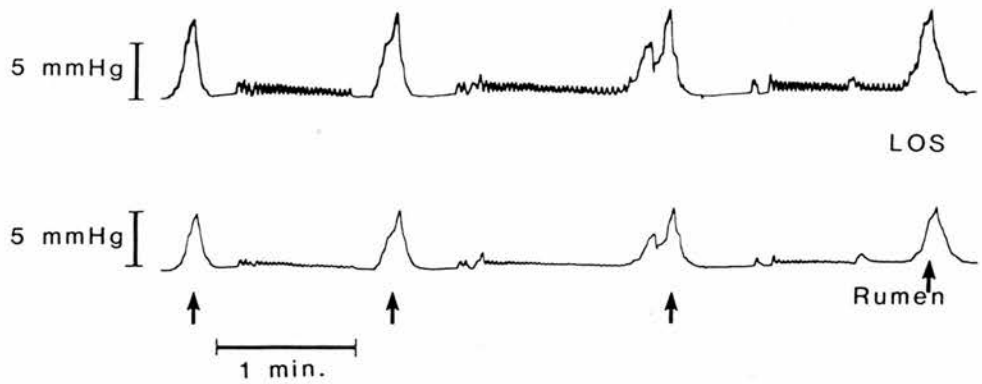


FIGURE 10.19

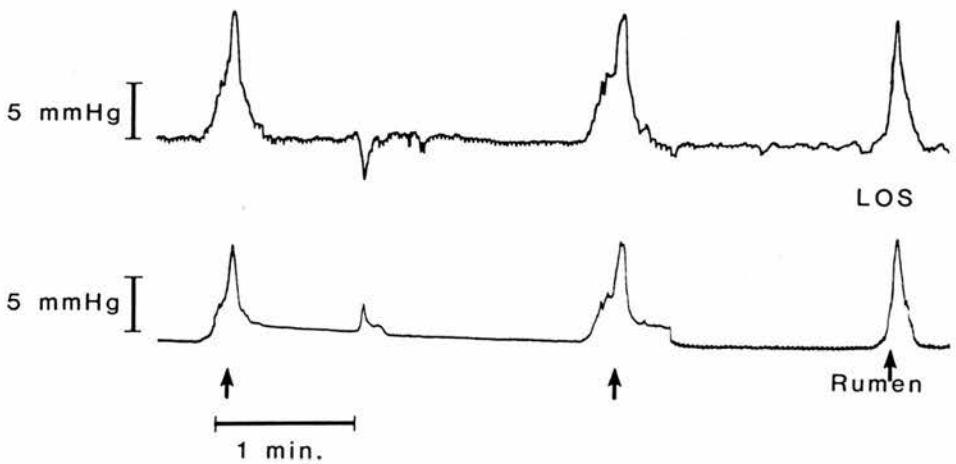


FIGURE 10.20

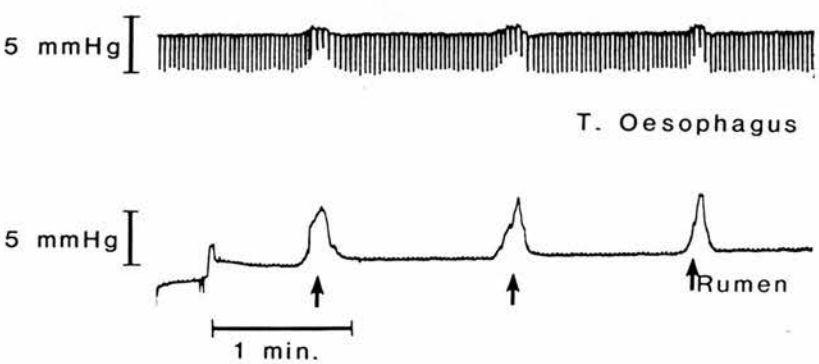


FIGURE 10.21

LOS and rumen pressure changes in response to balloon distension in the thoracic oesophagus in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). At the top horizontal bars, a 4.0 cm balloon was inflated with 25 cc air in the thoracic oesophagus (10.0 cm anterior to LOS). The LOS showed relaxation during balloon distension, but when a rumen contraction occurred during balloon distension, the LOS also had increased intraluminal pressure.

FIGURE 10.22

LOS and rumen pressure changes in response to oesophageal balloon distension in deep anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). At the top horizontal bars, a 4.0 cm balloon was inflated with 25 cc air in the thoracic oesophagus (10.0 cm anterior to LOS). The LOS pressure was increased associated with balloon distension.

FIGURE 10.21

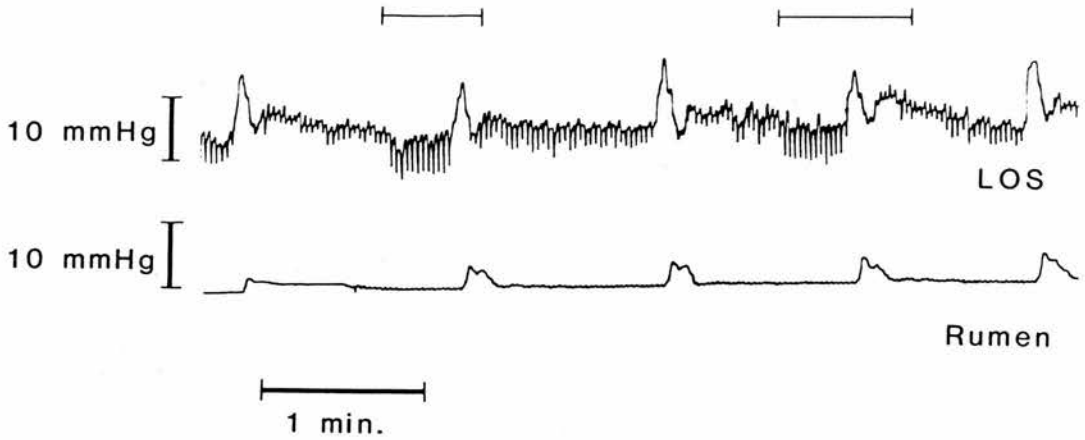
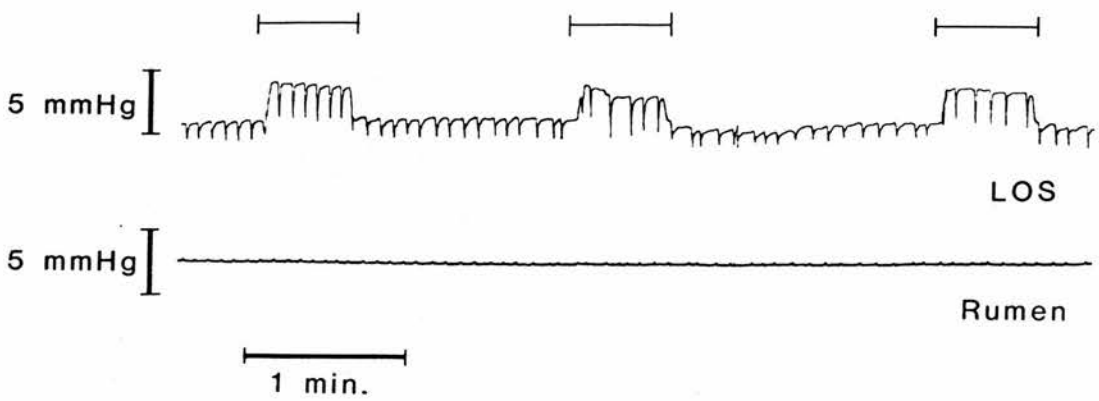


FIGURE 10.22



When a balloon was distended in the cervical oesophagus, the LOS tone was increased (in about 70% of cases) in both light and deep planes of anaesthesia (Figures 10.23 and 10.24). The oesophageal balloon distension inhibited respiratory pressure changes in about 70% of cases (Figure 10.25).

During the course of this investigation, great resistance was occasionally encountered in passing the catheter assembly down into the rumen and on several occasions, all attempts were unsuccessful in passing the tube across the LOS.

Discussion

This study has demonstrated for the first time the existence of a high pressure zone at the gastro-oesophageal junction which may act as a lower oesophageal sphincter (LOS). These results are in contrast to those reported by Winship et al. (1964) who were unable to record a high pressure zone in the sheep at this region using fluid filled open tip catheters and a "multiple pull through technique". In this technique, the catheter assembly is pulled from the stomach repeatedly in order to detect a high pressure zone at the lower oesophageal sphincter. This technique may be incapable of recording the high pressure zone at the lower oesophageal sphincter as the open tips have been reported to be blocked by the sphincteric mucosa (Pope, 1967; Winans and Harris, 1967). Winship et al. (1964) reported that a pressure gradient exists between the reticulorumen and oesophagus in sheep, but how this gradient was maintained in the absence of a LOS was not explained. In such a condition, the positive pressure in the reticulorumen is likely to be transmitted into the thoracic

FIGURE 10.23

LOS and rumen pressure changes in response to balloon distension in the cervical oesophagus in light anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). At the top horizontal bars, a 4.0 cm balloon was inflated with 25 cc air in the cervical oesophagus (10.0 cm anterior to thoracic inlet). The LOS showed an increase in tone in response to balloon distension. The rumen contractions did not correspond with balloon distension.

FIGURE 10.24

LOS and rumen pressure changes in response to balloon distension in the cervical oesophagus in deep anaesthesia (OTC). Upper tracing - LOS; lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). At the top horizontal bars, a 4.0 cm balloon was inflated with 25 cc air in the cervical oesophagus (10.0 cm anterior to thoracic inlet). The LOS showed an increase in tone associated with balloon distension. The rumen did not show any provoked activity.

FIGURE 10.25

Thoracic oesophageal and LOS pressure changes in response to balloon distension in the cervical oesophagus in light anaesthesia (OTC). Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - LOS. At the top horizontal bars, a 4.0 cm balloon was inflated with 25 cc air in the cervical oesophagus (10.0 cm anterior to thoracic inlet). The absence of respiratory pressure changes associated with balloon distensions indicates cessation of breathing. Neither tracings showed any marked change in tone during the distensions.

FIGURE 10.23

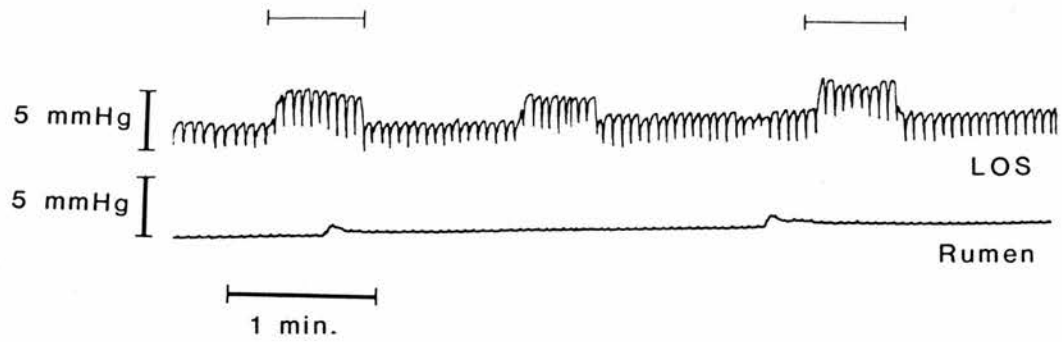


FIGURE 10.24

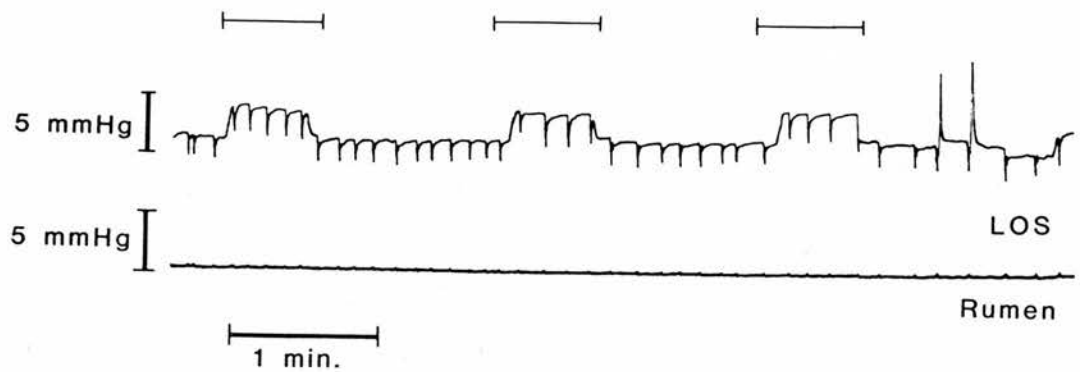
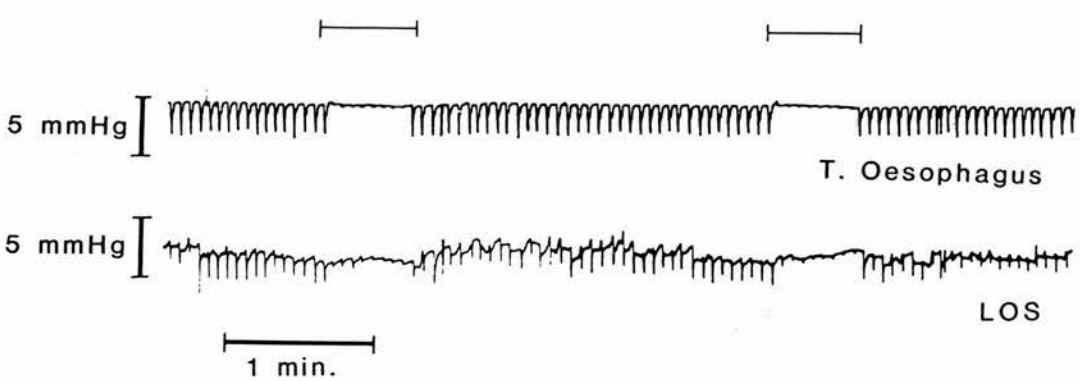


FIGURE 10.25



oesophagus and as a result maintenance of a pressure gradient across the LOS seems difficult.

The length of LOS was measured using a balloon tip catheter and a slow continuous pull through technique. The size of balloon is likely to influence the length of high pressure zone measured. The use of a longer balloon may magnify the exact length of this zone whereas a shorter balloon may have different sensitivities. These variables, however, were not investigated in the present studies. An accurate measurement of LOS length is probably not possible by this form of manometry. The length of LOS in the anaesthetized sheep using a 1.0 cm balloon was 2.9 cm which is comparable to that in man (2.6 cm) as reported by Botha et al. (1957).

On a few occasions, a two-stage high pressure zone was recorded at the gastro-oesophageal junction. Dougherty and Meredith (1955) demonstrated radiographically in conscious sheep that two sphincters are present in the lower oesophagus, one at the level of the diaphragm and the other one about 5.0 cm cranial to the diaphragm. These studies, however, did not involve simultaneous manometry. The demonstration of two areas of high pressure is interesting and deserves further investigation.

The present investigation demonstrates the functional pattern of LOS with respect to ruminal and oesophageal pressure changes. The LOS usually contracted just prior to rumen contraction and the pressure recorded usually had a greater amplitude (3-4 times the amplitude of rumen contractions). This pattern of LOS response may function to combat the higher intraruminal pressure and prevent oesophageal reflux. A gradient of pressure between the LOS and rumen is therefore maintained.

The close association between the intraluminal pressure changes in the LOS and rumen suggests that these two areas are functionally closely related. The ruminal and LOS contractions, however, did not influence the oesophageal pressure changes. Similar patterns of LOS contractions in response to increased intragastric pressure due to gastric contractions have also been reported in monogastric animals (Carlson, Boyd and Pearcy, 1922; Clark and Vane, 1961; Diamant and Akin, 1972). Relaxation of the stomach is also associated with the sphincteric relaxation (Burget and Zeller, 1936). The neural pathways for mediation of a sphincter contraction in association with a rumen contraction are still obscure. In monogastric animals, however, three possible mechanisms have been suggested (Carlson and Luckhardt, 1914; Carlson et al., 1922). These are (i) extension by local pathways in the myenteric plexus from the stomach, (ii) simultaneous motor impulses via the vagus to both stomach and LOS, (iii) vagally mediated motor control. The involvement of vagus in maintaining the LOS pressure has also been studied by Diamant and Akin (1972): bilateral vagotomy 1.0 cm above LOS abolished or reduced the LOS pressure in the dog. This suggests the presence of a vagal afferent or efferent either in the local myenteric plexus or mediated through a long vagal reflex.

The increase of LOS pressure in response to rumen contractions is not passive i.e. the LOS contraction is not a reflection of the rumen contraction but rather represents an active component, the amplitude of LOS contraction being greater than that of rumen contraction.

The results of control experiments showed that there was no significant difference between the amount of increased intraruminal pressure and that of LOS pressure. This suggests that the LOS contraction in response to increased intraruminal pressure due to rumen contraction is more forceful than that due to abdominal compression. This indicates the involvement of a different mechanism of LOS contraction in the case of abdominal compression.

Intraluminal balloon distension in the thoracic oesophagus in light anaesthesia caused relaxation of LOS, while in deep anaesthesia the sphincteric tone was increased. Similar results have been reported in conscious humans (Creamer and Sehlegel, 1957). Code et al. (1958) suggested that the LOS relaxation in man associated with balloon distension may be due to vagally mediated reflex mechanisms. When a rumen contraction occurred, the LOS contracted even when it was relatively relaxed due to balloon distension. Possible explanations of this event are: (i) a central discharge of motor impulses to the LOS and rumen which dominates over the inhibitory effects caused by oesophageal balloon distension; (ii) a local reflex mediated through myenteric plexus; (iii) a local hormone which may be produced in association with rumen contractions. The mechanism responsible for an increase in LOS tone during balloon distension in deep anaesthesia is not understood.

In the present investigation, intraluminal distension of a balloon in the oesophagus frequently caused cessation of breathing. Troyer and Rosso (1982) also reported similar results in the anaesthetized dog and this is probably associated with a reflex which is elicited by the distension of the oesophagus and inhibits selectively the diaphragm

via the vagal afferent nerves. This inhibition of phrenic motoneuron may produce apnoea.

No single factor appears to be responsible for maintaining LOS tone, but probably it involves a number of co-ordinated factors which may include (i) a tonic excitatory nerve activity of either extrinsic or intrinsic origin, (ii) a circulating gastro-intestinal hormone, (iii) a local hormone, (iv) inherent properties of the smooth muscle cell, (v) a combination of two or more of the above factors (Goyal and Cobb, 1981).

It is sometimes assumed that the intramural innervation of the LOS is more dense than that of other parts of the oesophagus. Christensen and Robinson (1982) however, demonstrated in the opossum that the existence of intramural plexus in the LOS areas is less, suggesting that the intrinsic innervation may not be greatly involved in maintaining the LOS tone.

Great difficulty was sometimes encountered in passing the catheter assembly down into the rumen. On several occasions, all attempts failed to insert the tube down to the rumen. This problem has also been encountered in the dog by Rosin, Galphine and Bowen (1979). There is very little information about this problem except in the conscious state where the LOS relaxes following swallowing. The presence of the catheter along the oesophagus, moreover, excites swallowing and therefore the insertion of the catheter assembly is less problematical in the conscious state. On the other hand, under the conditions reported here, the LOS tends to maintain a relatively uniform pressure since the swallowing reflex is abolished in the relatively deeply anaesthetized sheep.

This constant barrier of LOS perhaps offers resistance to catheter insertion. The other factor may be the anatomical relationship of the ruminant digestive tract. In one sheep during its last anaesthetic, a laparotomy was performed and a stomach tube was introduced along the oesophagus and strong resistance was observed. On palpation, the tip of the stomach tube was found to be stuck against the diaphragm on each occasion the tube was advanced; but when the tube was held properly by manipulation through the abdominal wound, it entered the rumen with relatively little resistance. In ruminants, the oesophagus makes an angle with the reticulorumen. Also, the organ remains flaccid at rest while the LOS maintains continuous tone and remains tonically contracted. Moreover, the caudal part of the oesophagus is widely dilated in comparison with the cranial parts (Sisson and Grossman, 1975). In this situation, when a stomach tube is advanced through the oesophagus, it may not follow the true course of the organ and become stuck against the diaphragm. This problem was more frequently observed in the unstarved sheep with head down position. In such a position, the reticulorumen compresses the diaphragm cranially and may make the oesophageal ruminal angle more acute. The insertion of the tube may be more easily achieved if the hind part of the animal is tilted down. In this case, the degree of the above angle may be reduced and also the oesophagus may be relatively stretched due to downward traction by the reticulorumen. Therefore, the difficulty of catheter insertion may be in part associated with the anatomical relationships as well as to tonicity of LOS muscle.

The resting pressure in the rumen is always in excess of that in the oesophagus. So, if there is no barrier between the two organs, reflux should occur from the reticulorumen into the oesophagus. The presence of LOS in the anaesthetized sheep probably acts as this barrier. In humans also, this barrier has been reported to be maintained by the tonic contraction of the LOS circular muscle (Fyke et al., 1956).

Several mechanisms may be responsible for maintaining gastro-oesophageal competence. The anti-reflux mechanism in the dog is mainly due to intrinsic LOS tone (Laitinen et al., 1978) although a significant role by mechanical factors has been forwarded by Dodds et al. (1976). Diamant and Akin (1972) reported in the unanaesthetized dog that under those circumstances which favour reflux, the LOS acts as a barrier to prevent or minimize its occurrence.

The anatomical location of LOS in the ruminant has not been studied. There are some reports in monogastric animals, but these appear to be confusing as to whether it is pre-diaphragmatic, post-diaphragmatic or at the level of the diaphragm (Wankling et al., 1965; Lind et al., 1966; Skinner and Camp, 1968; Rayl et al., 1972). In the present investigation, the interpretation of the withdrawal tracings suggests that the LOS in sheep lies just posterior to or at the level of the diaphragm and not cranial to that, in which case the withdrawal tracing should record first a positive intraruminal pressure, then a negative intrathoracic pressure, then the high pressure zone and again a negative pressure. Radiographic methods are also reported to be unsatisfactory in identifying the LOS since swallows of radio-opaque or oesophageal placement of radio-opaque

material causes relaxation and opening of the LOS (Goyal and Cobb, 1981). An exact location of the high pressure zone, therefore, remains unclear.

There are no reports in the literature describing continuous long term changes in LOS motor activity in ruminants. In the present study, it was possible to observe simultaneously changes of LOS and rumen in anaesthetized sheep during periods of up to one hour. The traditional pull through technique measures the LOS pressure for only a moment and is not suitable for monitoring continuous changes. In the present study, a "stationary catheter technique" was employed which is also useful to record the pattern of LOS response to different stimuli. In conscious animals, however, this technique may be impracticable. The major disadvantage is that the position of the catheter may be displaced from time to time. Therefore, attention to the tracing is essential to reposition the catheter as soon as it is displaced.

The continuous monitoring of LOS pressure shows that the base line pressure is not constant. The resting pressure shows rhythmic variations. The most prominent pressure fluctuations are those related to respiratory excursions and rumen contractions. The fluctuation of baseline pressure may be to some extent due to forward and backward movement of the LOS associated with respiration (Winans, 1972).

Most of the present manometric devices involve insertion of catheter or catheter assembly within the lumen of the sphincter to measure its pressure. This stretches the sphincter and modifies the true resting state since the vagal mechanoreceptors of the LOS in

sheep are very sensitive to mechanical distension (Falempin et al., 1978).

Conclusions

(i) A zone of high pressure is present at the junction of the oesophagus and reticulorumen, i.e. these studies confirm the existence of LOS in the anaesthetized sheep.

(ii) The LOS is involved in the regulation of gastro-oesophageal reflux.

(iii) The tone of the LOS was not influenced by the depth of anaesthesia in these experiments.

(iv) The position of LOS may be just posterior to the diaphragm, and may consist of more than one high pressure zone.

CHAPTER ELEVEN

THE INFLUENCE OF PENTAGASTRIN, ATROPINE AND PROPRANOLOL ON LOWER OESOPHAGEAL SPHINCTER PRESSURE

Introduction

The lower oesophageal sphincter (LOS) has been demonstrated to be an important component in controlling gastro-oesophageal reflux in anaesthetized sheep (Chapter 10). It would appear that the LOS performs this function by the tonic contraction of its muscle. The factors which influence the strength of LOS, might therefore, influence the occurrence of reflux. These factors are mainly nervous and humoral. The gastro-intestinal hormones, particularly gastrin, which is secreted by the gastric antrum is widely reported to stimulate the LOS tone in man (Giles et al., 1969; Lipshutz and Cohen, 1971). This hormone is also present in the sheep and is secreted from the pyloric and fundic regions of the abomasum (Anderson, Fletcher, Pitts and Harkins, 1962). But there is apparently no information as to whether this hormone augments the sphincter pressure in ruminants during anaesthesia.

It is reported that bilateral stimulation of cervical vagi in sheep causes tonic closure of LOS (Habel, 1956). Atropine is a drug which is used predominantly in monogastric animals as a premedicant. Bettarello, Tuttle and Grossman (1960) reported that atropine increases the risk of gastro-oesophageal reflux in man by lowering the sphincteric tone. This action of atropine on ruminant LOS has not however been tested.

The nervous control of LOS is still unclear and the results are conflicting (Chapter 2). The reports of the influence of sympathetic nerves on LOS pressure are limited. Experiments were performed in the anaesthetized sheep to study the effects of pentagastrin, atropine and propranolol on lower oesophageal sphincter pressure.

Materials and Methods

Ten starved sheep were used in this investigation. Induction of anaesthesia was achieved with halothane (4%), nitrous oxide and oxygen (50:50) using a face mask and Magill system and maintained in a similar fashion as described in the general methods (Chapter 4). The sheep were positioned in right lateral recumbancy during anaesthesia. The LOS pressure was measured with balloon tip catheter (1.0 cm long balloon with 0.2-0.3 cc air) using catheter assembly 1 (Chapter 4) over a period of 60 minutes.

After intubating the trachea, the injection site (recurrent tarsal vein) was clipped and sprayed with chlorehexidine gluconate (Hibitane , ICI Ltd., Great Britain).

Pentagastrin, atropine and propranolol were used at the following dose rates:

Pentagastrin (Peptavlon, ICI Ltd., Great Britain):
6.0 $\mu g.kg^{-1}$, 12.0 $\mu g.kg^{-1}$, 24.0 $\mu g.kg^{-1}$

Atropine sulphate (Phoenix Pharmaceuticals Ltd., U.K.):
15.0 $\mu g.kg^{-1}$, 30.0 $\mu g.kg^{-1}$

Propranolol hydrochloride (Inderal, ICI Ltd., Great Britain):
0.05 $\mu g.kg^{-1}$

These drugs were administered as a single bolus injection via the recurrent tarsal vein. In any one anaesthetic, only a single dose of one of these drugs was administered. A control dose of saline was also administered by the same route 30 minutes prior to the test dose except in the case of atropine. The LOS pressures were measured for two equal periods i.e. up to 30 minutes after saline injection and up to another 30 minutes after the administration of

the drugs. In the case of propranolol, atropine (30.0 $\mu g.kg^{-1}$) was injected intravenously five minutes prior to the administration of the drug to minimize the vagal suppression of the heart associated with propranolol.

The data obtained were analysed using the Student's t-test.

Results

Pentagastrin: Intravenous administration of pentagastrin produced an increase in LOS pressure within one minute of injection (Figure 11.1) in the majority of cases. Similar injections with equivalent doses of saline did not produce any increase in LOS pressure. These pressures continued to be increased gradually until the end of 30 minutes. The LOS pressure values recorded after saline and pentagastrin injections are presented in Appendices 11.1-11.3. The LOS pressures recorded after pentagastrin for different dose groups did not differ significantly when compared with the respective saline values. A representative picture of the mean LOS pressures after saline and pentagastrin (24.0 $\mu g.kg^{-1}$) is presented in Figure 11.2. The responses of LOS to different doses of pentagastrin (6.0 $\mu g.kg^{-1}$, 12.0 $\mu g.kg^{-1}$, 24.0 $\mu g.kg^{-1}$) are presented in Figure 11.3. These differences were again not significant.

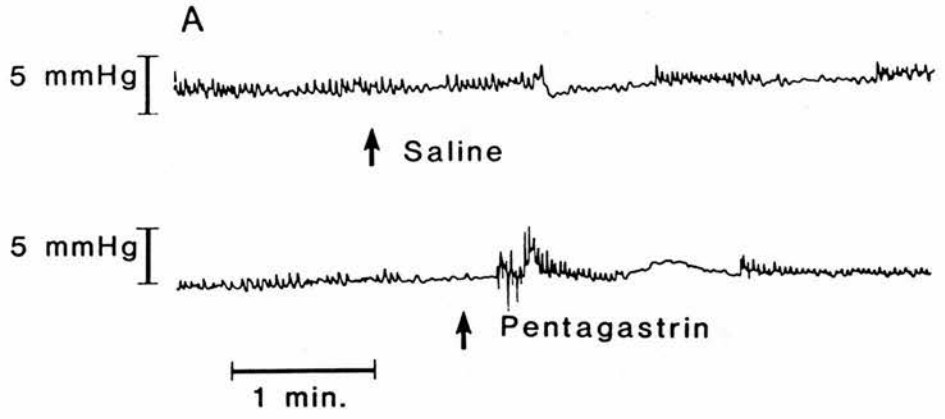
Atropine sulphate: The LOS pressure was slightly decreased within one minute of the intravenous administration of atropine (Figure 11.4). The individual records of this pressure for the two different doses of atropine (15.0 $\mu g.kg^{-1}$ and 30.0 $\mu g.kg^{-1}$) are presented in Appendices 11.4 and 11.5. There was no significant difference in LOS pressure between the two dose groups when compared with

FIGURE 11.1

A. Effect of pentagastrin (6.0 $\mu g.kg^{-1}$) on lower oesophageal sphincter (LOS) pressure. Upper tracing - LOS pressure changes showing the influence of saline; lower tracing - LOS pressure changes showing the influence of pentagastrin. The arrows show the timing of the administration of saline and pentagastrin. The LOS did not show any change in pressure after saline. Pentagastrin produced an increase in pressure within one minute of injection.

B. Effect of pentagastrin (24.0 $\mu g.kg^{-1}$) on LOS pressure. Upper tracing - LOS pressure changes showing the influence of saline; lower tracing - LOS pressure changes showing the influence of pentagastrin. The arrows show the timing of the administration of saline and pentagastrin. The LOS did not show any change in pressure after saline but pentagastrin produced an increase in pressure within one minute of injection. The abrupt change in pressure (*) probably relates to rumen contraction.

FIGURE 11.1


A
5 mmHg
Saline
5 mmHg
Pentagastrin
1 min.

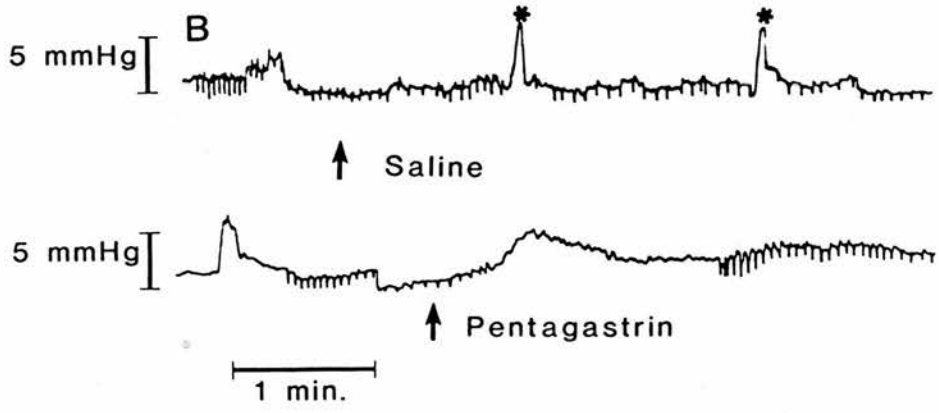

5 mmHg
B
Saline
5 mmHg
Pentagastrin
1 min.

FIGURE 11.2

Effect of intravenous bolus injection of pentagastrin (24.0 $\mu g.kg^{-1}$) on lower oesophageal sphincter pressure (n=5).The baseline (100) represents a mean value of intraluminal pressure measurements of LOS made in the 5 minutes prior to injection of pentagastrin or saline at 0 minutes.(No significant difference).

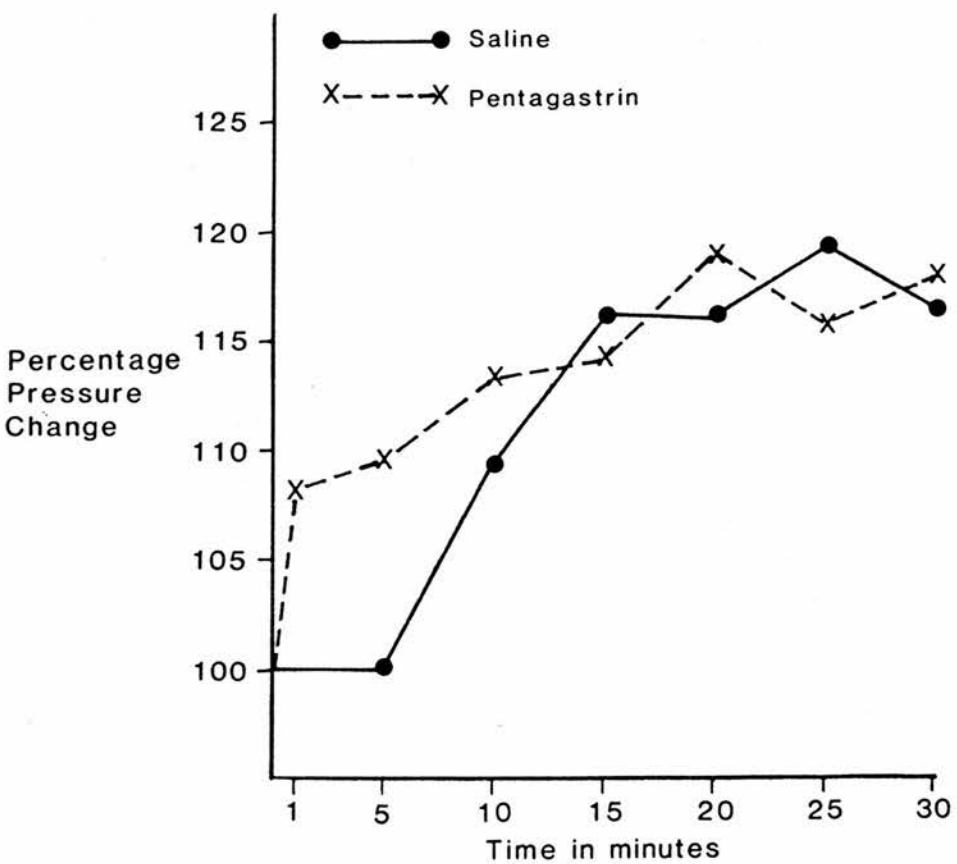


FIGURE 11.3
Pooled data demonstrating the effects of intravenous bolus injections of pentragastrin (6.0 μg.kg^{-1}, 12.0μg.kg^{-1}, and 24.0μg.kg^{-1}) on lower oesophageal sphincter(LOS) pressure.(No significant difference).

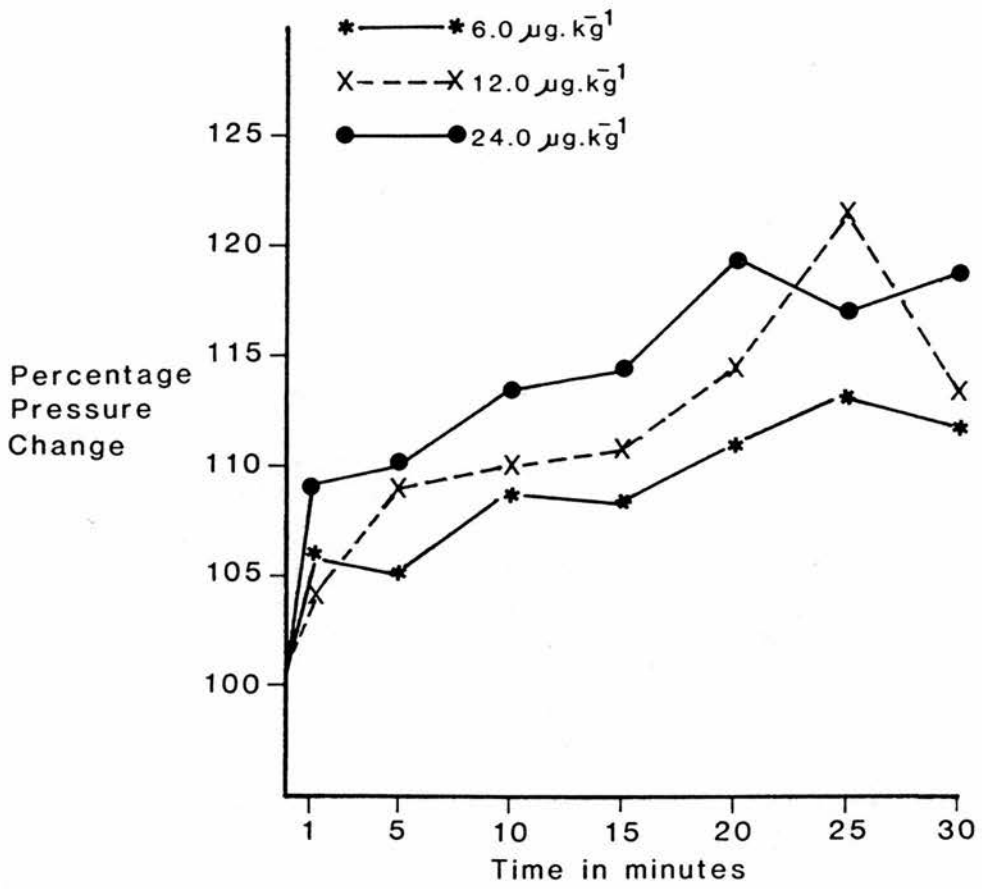


FIGURE 11.4

A. Effect of atropine (15.0 $\mu g.kg^{-1}$) on LOS pressure. The arrow shows the timing of atropine injection. The LOS pressure was decreased for a short time within one minute of injection before returning to control values.

B. Effect of atropine (30.0 $\mu g.kg^{-1}$) on LOS pressure. Upper tracing - LOS pressure changes before the administration of atropine; lower tracing - LOS pressure changes showing the influence of atropine. The arrow indicates the timing of atropine injection. The LOS pressure was decreased within one minute of injection and returned to baseline levels after two minutes. The pressure waves (*) probably were associated with rumen contraction.

FIGURE 11.7

Effect of propranolol hydrochloride (0.05 $\mu g.kg^{-1}$) on LOS pressure changes. Upper tracing - LOS pressure changes showing the influence of saline; lower tracing - LOS pressure changes showing the influence of propranolol. The arrows show the timing of the administration of saline and propranolol. In the case of propranolol, the animal was atropinized (30.0 $\mu g.kg^{-1}$) 5 minutes prior to the administration of the drug. The LOS did not show any change in pressure after saline. The pressure waves present during saline control (*) probably relate to reticuloruminal contractions. These pressure waves, however, were suppressed after atropine and propranolol injections for a variable length of time.

FIGURE 11.4

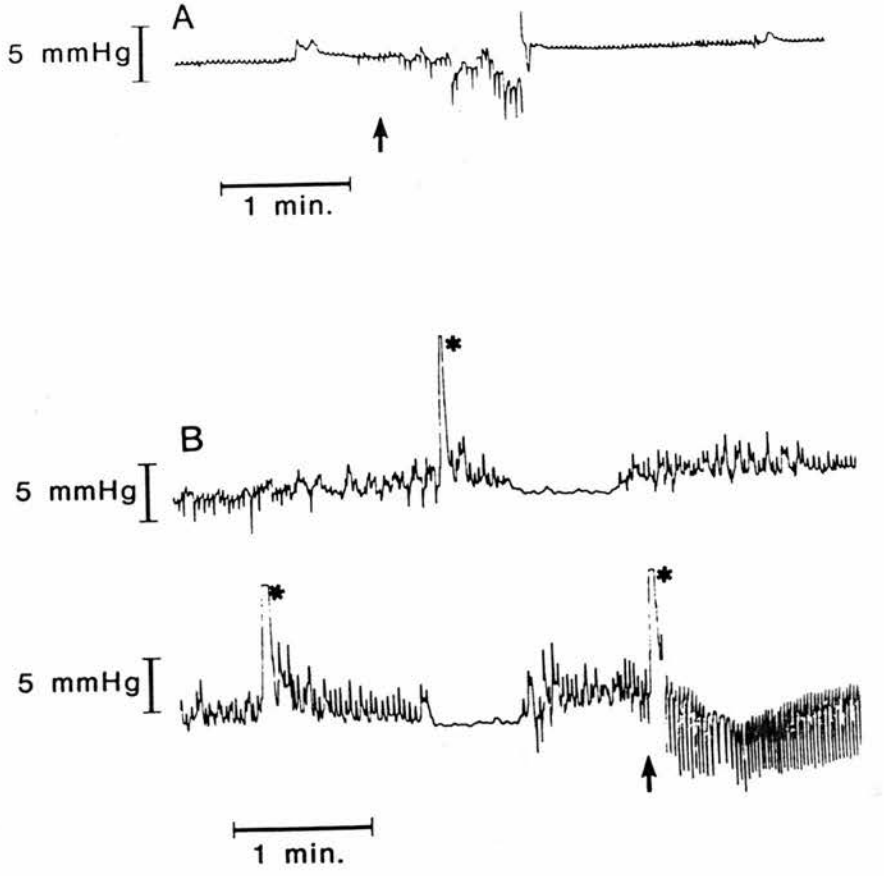
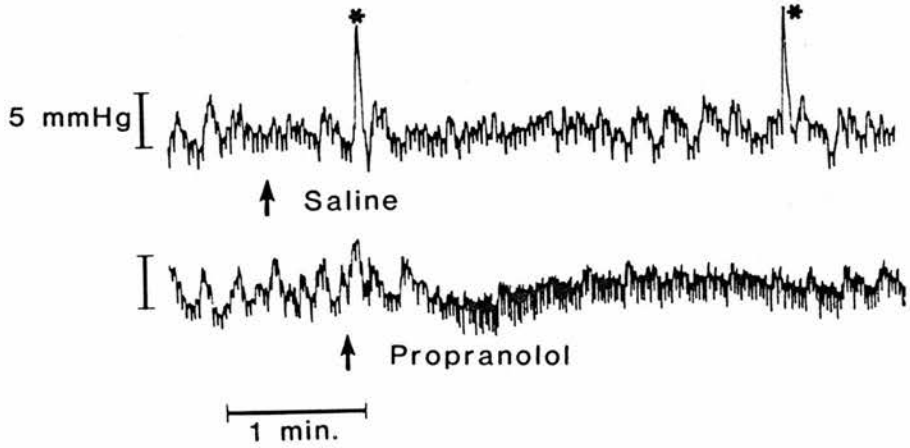


FIGURE 11.7



the basal pressures (Figure 11.5) although the LOS pressures were relatively more decreased with the higher dose (30 $\mu g.kg^{-1}$), but there was no significant difference between the two dose groups.

Usually, following atropinization, the LOS tone decreased within one minute and the characteristic LOS pressure changes were suppressed for a variable length of time. With the higher dose (30 $\mu g.kg^{-1}$) this suppression sometimes continued up to 30 minutes post injection.

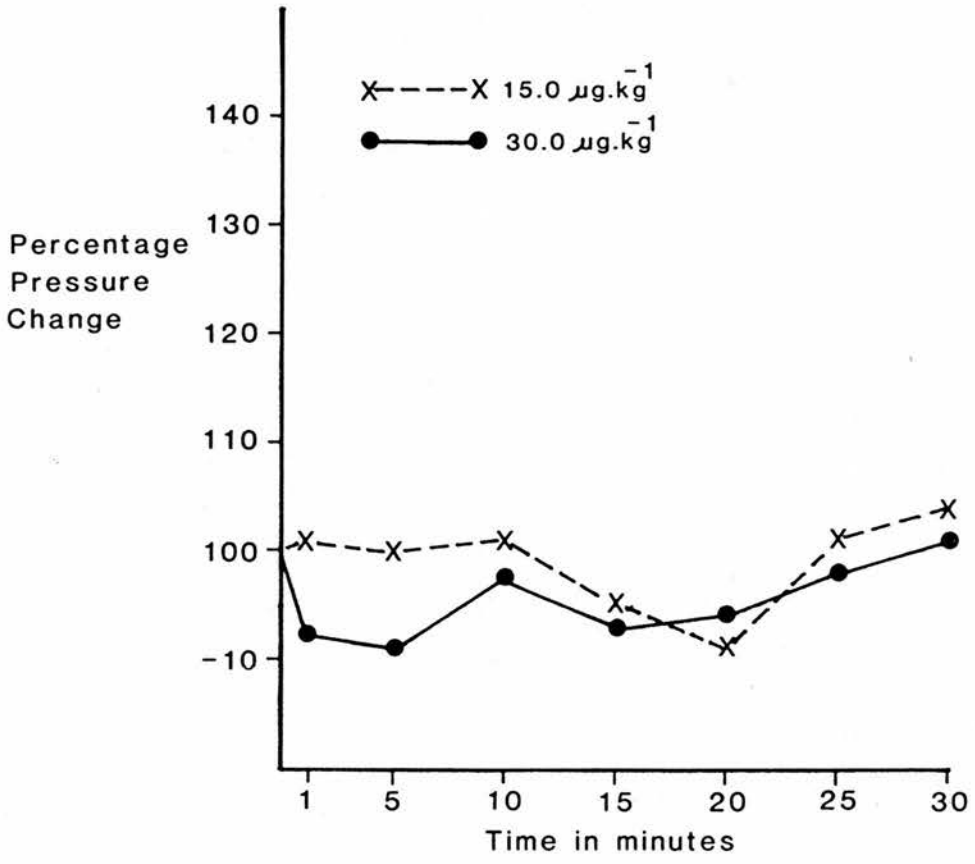
Propranolol hydrochloride (after atropine): The influence of this drug on LOS pressure is presented in Appendix 11.6. After propranolol, the intraluminal pressure of LOS gradually decreased (Figure 11.6). There was, however, no significant difference when compared with the control values. The LOS pressure was relatively decreased or underwent no changes following propranolol injection but the pattern of LOS pressure changes was modified (Figure 11.7). The LOS after the dose of propranolol did not show larger pressure waves as were seen after saline control injections.

In the present investigation, the intraluminal pressures of LOS during saline control were different (Figures 11.2 and 11.6) although this difference was not statistically significant.

Discussion

In these series of experiments, starved sheep were used. The idea was to keep the rumen pressure steadier so that the sphincter pressure does not increase with the increase of intraruminal pressure as is the case with the unstarved animals. It makes it difficult to identify the actual change in LOS pressure relating to drugs.

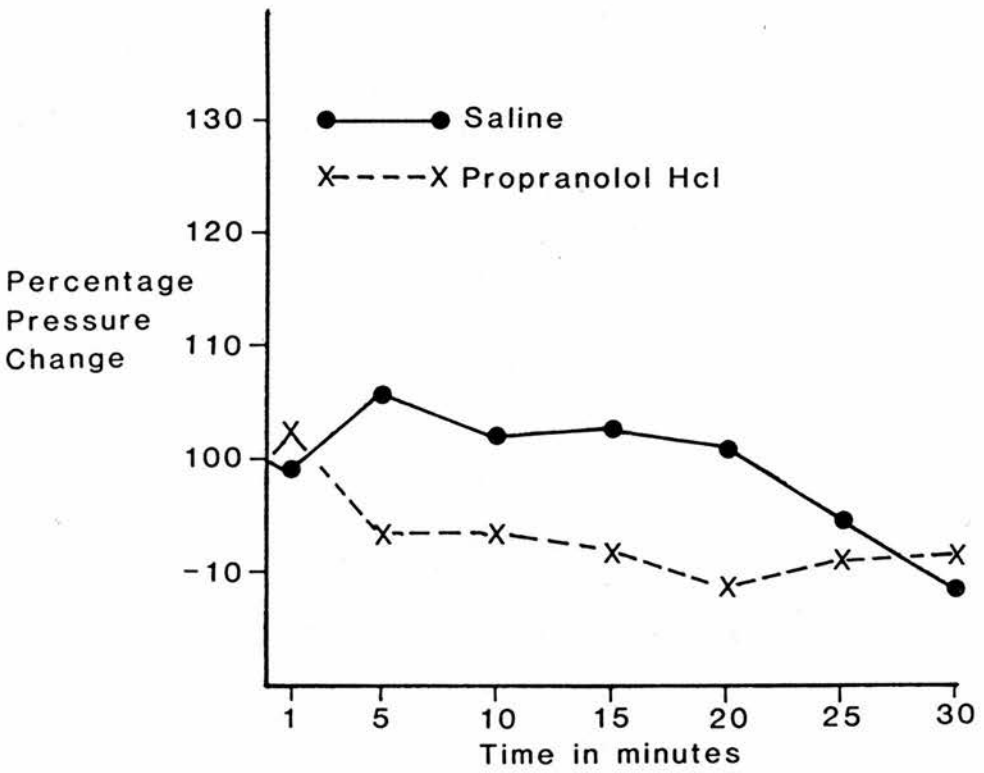
FIGURE 11.5
Pooled data demonstrating the effects of intravenous bolus injection of atropine sulphate (15.0 $\mu g.kg^{-1}$, and 30.0 $\mu g.kg^{-1}$) on lower oesophageal sphincter pressure.



(No significant difference)

FIGURE 11.6

Effect of intravenous bolus injection of propranolol Hcl (0.05 $\mu g.kg^{-1}$) on lower oesophageal sphincter pressure.



(No significant difference)

The regulation of lower oesophageal sphincter (LOS) in ruminants is not clearly understood. The gastro-intestinal hormones particularly gastrin have been reported to stimulate the LOS tone in man (Giles et al., 1969; Lipshutz and Cohen, 1971). The present studies in anaesthetized sheep, however, failed to demonstrate any significant influence of pentagastrin on LOS pressure although the drug caused a variable increase in LOS tone. This drug is very similar to gastrin and has been widely used as a substitute for gastrin in different monogastric animals and man to investigate its influence on LOS pressure. Despite a great deal of study, there still remains considerable confusion about whether the action of gastrin is physiological or pharmacological (Grossman, 1973; Jensen et al., 1980). Two guidelines are usually used in ascertaining whether an effect is physiological (Johnson, 1974). Firstly, the effect should take place in response to a dose of the hormone which does not exceed the maximal dose for the primary action, e.g. for gastrin the primary action is gastric acid secretion. Secondly, the effect should be produced by endogenous release of the hormone, e.g. endogenous release of gastrin occurs with topical application of acetylcholine to the gastric antrum (Johnson, 1974). There is impressive evidence for the physiological role of gastrin in relation to the LOS in the opossum (Lipshutz and Cohen, 1971), but in the dog peak sphincter pressure responses and peak gastrin levels after meals do not coincide in time (Hollenbeck et al., 1975). In the present investigation, the maximum LOS pressure was recorded with a lower dose of pentagastrin (12.0 $\mu g.kg^{-1}$) which is indirect evidence for the possible physiological action of gastrin. In the case of pharmacological action,

the highest response should be recorded with the greater dose. Alternatively, the lowest doses may have been above the physiological range.

In the present studies, pentagastrin caused a transient increase in LOS pressure within one minute of injection in 80% of cases and then maintained a very slow increase often indistinguishable from saline control. In humans, the LOS pressures begin to rise within two minutes of subcutaneous injection of pentagastrin (1.0 $\mu g.kg^{-1}$), the peak is attained in seven minutes and returned to pre-injection levels by 30 minutes (Castell and Harris, 1970). The transient action of pentagastrin on LOS may be due to rapid elimination of the hormone by the liver into the bile (Stagg, Temperley and Wyllie, 1971).

The doses of hormone used in the present studies were much higher than those used in the dog (McGuigan et al., 1971). The doses administered as a continuous perfusion were 0.125, 0.250, 0.50, 1.0 and 2.0 $\mu g.kg^{-1}$/hour in contrast to bolus doses used in the present studies (6.0 $\mu g.kg^{-1}$, 12.0 $\mu g.kg^{-1}$ and 24.0 $\mu g.kg^{-1}$ intravenously). Ariens (reviewed by Cohen and Lipshutz, 1971) postulated that the increased dose of gastrin cannot exert its action on LOS because of auto inhibition which is thought to be due to saturation of all high affinity excitatory receptors with additional gastrin molecules combining with a low affinity inhibitory receptor. Similar views were expressed by Zwick, Bowes, Daniel and Sarna (1976) who demonstrated in the anaesthetized dog that an intravenous dose of pentagastrin (3.0 $\mu g.kg^{-1}$) produced peak sphincter pressure whereas higher doses (6.0 $ug.kg^{-1}$) in the same route produced a lesser effect of a short duration and concluded that the stimulatory effect of pentagastrin is mainly due

to a direct action on the LOS. Another possibility is that the stimulatory effect of pentogastrin is not directly on the smooth muscle but on receptors in nerves that are not atropine sensitive. Zwick et al. (1976) suggested that large doses of pentagastrin have both stimulatory and inhibitory effects and that the inhibitory effect is mediated partly via preganglionic neurons acting through adrenergic receptors and partly through beta adrenergic receptors.

In monogastric animals, exogenous gastrin injection induces gastric acid secretion which changes the gastric pH. The contact of these gastric contents with the LOS may be responsible for the increase of sphincter pressure (Giles et al., 1969). In ruminants, however, gastric acid is produced in the abomasum and not in the rumen and as a result there is no contact of gastric contents (abomasal contents) with the LOS. Therefore, the effect of gastrin in elevating the LOS tone may be due to the direct action of gastrin on LOS muscles.

During anaesthesia, the rumen gets distended due to cessation of eructation and continuous production of gases. This ruminal distension may stimulate the release of endogenous gastrin by a non-cholinergic mechanism (Schiller, Walsh and Feldman, 1980) which in turn may be involved in maintaining the increased tone of the sphincter and prevents reflux. The tone of LOS, however, may occasionally be relaxed during anaesthesia and reflux occurs. This sphincteric relaxation may be associated with the endogenous secretion of some other gastrointestinal hormones, e.g. secretin, prostaglandins. These hormones have been reported to be inhibitory to the

LOS tone (Hansky, Soveny and Korman, 1971; Goyal et al., 1973). Hanksy et al. (1971) reported that the intravenous injection of secretin (1.0 $U.kg^{-1}$) produced a significant fall in serum gastrin from a mean fasting level of 60 pg/ml to 15 pg/ml at a mean time of 25 minutes after injection. They concluded that secretin may act not only as an inhibitor of gastrin action at a receptor site, but also as a suppressor of gastrin release.

In the present studies, atropine produced little decrease in LOS tone. The vagus nerve is involved in maintaining the resting LOS pressure (Roman and Gonella, 1981). Atropine is a parasympatholytic drug, i.e. it abolishes the activities of vagus nerves by blocking the effects of acetylcholine at muscarinic post-ganglionic parasympathetic nerve endings. Therefore, this drug is likely to produce inhibition to LOS tone. Gonella, Niel and Roman (1979) reported in the cat that atropine may also antagonize the motor effects of the sympathetic system on LOS.

The majority of the workers accept that atropinization in man is associated with diminished LOS pressure and therefore more vulnerable to reflux (Lind et al., 1968; Skinner and Camp, 1968; Hall et al., 1975). The present investigation shows that atropine has an insignificant action on LOS pressure at the doses used in anaesthetized sheep and was not associated with the increased occurrence of reflux.

The effects of atropine on LOS tone in man have been studied with conflicting results (Bettarello et al., 1960; Skinner and Camp, 1968). Skinner and Camp (1968) reported that atropine significantly increased the occurrence of reflux by diminishing the LOS pressure.

In the case of atropine, no saline control injections were administered to the experimental animals, but in a similar experiment with pentagastrin, the injection of saline did not produce any significant change in LOS pressure. This also agrees with the findings of Skinner and Camp (1968).

Propranolol is an adrenergic beta antagonist, i.e. it inhibits the adrenergic beta-receptors. Di Marino and Cohen (1975) reported that stimulation of beta-adrenoceptor reduces the tone of lower oesophageal sphincter (LOS) in the opossum. Therefore, theoretically the LOS tone should be increased with the inhibition of these receptors. Propranolol is a drug which inhibits the beta-receptors and as a result an increase in LOS tone is anticipated. In the present investigation, however, propranolol did not increase the LOS tone but rather decreased it. The reason for this decrease may be associated with the use of atropine as a premedicant. Propranolol is a cardioselective beta adrenoceptor blocking drug (Veter et al., 1982). This drug when administered may produce cardiac failure from the dominance of vagus nerves. Accordingly, atropine was injected prior to the administration of propranolol to cut down the function of vagus so that the heart function was not compromised. Therefore, in the present study, the action of propranolol (if any) may have been antagonized by atropine. Veter et al. (1982) however, reported that the human LOS is not sensitive to propranolol as the LOS tone was not significantly increased.

The B blocking efficacy of the dose of propranolol used (0.05 µg/kg) has not been established. It may well be that the dose employed was too small to have any significant B blocking activity.

From these results, the following general conclusions can be drawn:

(i) The gastrin analogue pentagastrin, at the dose levels used increases LOS pressure briefly.

(ii) Atropine sulphate decreases LOS pressure briefly.

(ii) Propranolol hydrochloride when administered after atropine caused no significant change in LOS pressure.

It is not possible to have much faith in the validity of these conclusions as the response to the saline control varied widely between sheep and also within the same sheep. Sheep 510D, for example, on two occasions showed a fall in LOS pressure following saline injection but on a third occasion a significant rise in pressure followed saline infusion.

CHAPTER TWELVE

AN ELECTROMYOGRAPHIC STUDY OF THE ACTIVITIES OF THE OESOPHAGUS, GASTRO-OESOPHAGEAL JUNCTION AND RETICULORUMEN

Introduction

Electromyography (EMG) is a technique which has frequently been employed to study the motor activities of the alimentary tract and other systems. Mechanical events produced by changes in muscular activity are associated with changes in the electrical status of the muscle fibres i.e. muscular contractions are associated with action potentials. Thus electromyographic activity is indicative of mechanical activity (Ruckebusch, 1970). The synchronization of EMG discharge with mechanical activity provides the most credible evidence for active motor function. There are few electromyographic studies of the functions of the oesophagus and the lower oesophageal sphincter in the anaesthetized ruminant. In the present investigation, electromyography from the oesophagus, gastro-oesophageal junction and reticulorumen were recorded in both conscious and anaesthetized sheep with the aim of characterizing the electrical activities of these organs and also to correlate them with the mechanical activities.

Materials and Methods

Four sheep prepared with chronically implanted electrodes were used in this study. The electrodes were made from nichrome wire (80% nickel, 20% chrome, Johnson Matthey Company, London). The diameter of the electrodes was 120 μ. The length of the electrodes depended on the anatomical demands of the sites to be implanted and was approximately 80 cm.

Each set of electrodes comprised of three wires which were ensheathed in portex vinyl tubing for protection and ease of handling.

The position of the electrodes inside the vinyl tubing was secured by tying both ends of the sheath with cotton thread and then sealed using a quick drying acrylic, M-coat D (Welwyn Strain Measurement Ltd.) (Figure 12.1).

At both ends of each electrode, the insulation was removed by flaming the wire at an approximate distance of 3.0 cm from the M-coat D sealing. The burned areas of the electrodes were further scratched with a blade to ensure that all insulation was removed.

Sites of Implantation

(i) Cervical oesophagus 5.0 cm anterior to the thoracic inlet.

(ii) Gastro-oesophageal junction 1.5 cm posterior to diaphragmatic hiatus.

(iii) Dorsal sac of rumen.

(iv) Reticulum.

Preparation of the Animal and Anaesthesia

The sheep were starved and water withheld for 24 hours. Induction of anaesthesia was performed using alphazalone/alphadolone (Saffan, Glaxovet Ltd.) and maintained using a gaseous mixture of halothane and nitrous oxide and oxygen (50:50) as described for the ruminal cannulation procedure (Chapter 8).

Implantation Technique

Introduction of the electrodes into the muscle was facilitated with curved 23-gauge hypodermic needles. The uninsulated part of the electrode was positioned in the muscle tissue and anchored in place by knotting as illustrated (Figure 12.2). The electrodes were

FIGURE 12.1

Electrodes ensheathed within vinyl tubing.

a. Vinyl tubing

b. Electrodes (nichrome wire)

c. M-coat D sealing

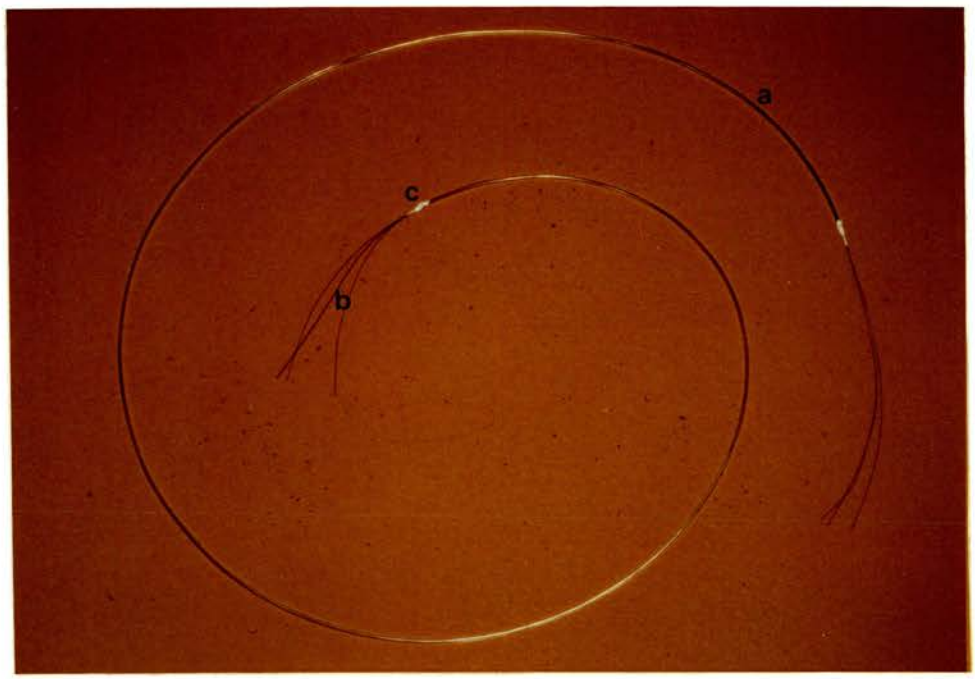

a
c
b

FIGURE 12.2

Technique of implantation of EMG electrodes.

(1) A curved hypodermic needle was passed through the muscular coat and the electrode to be implanted was inserted through the lumen of the needle.

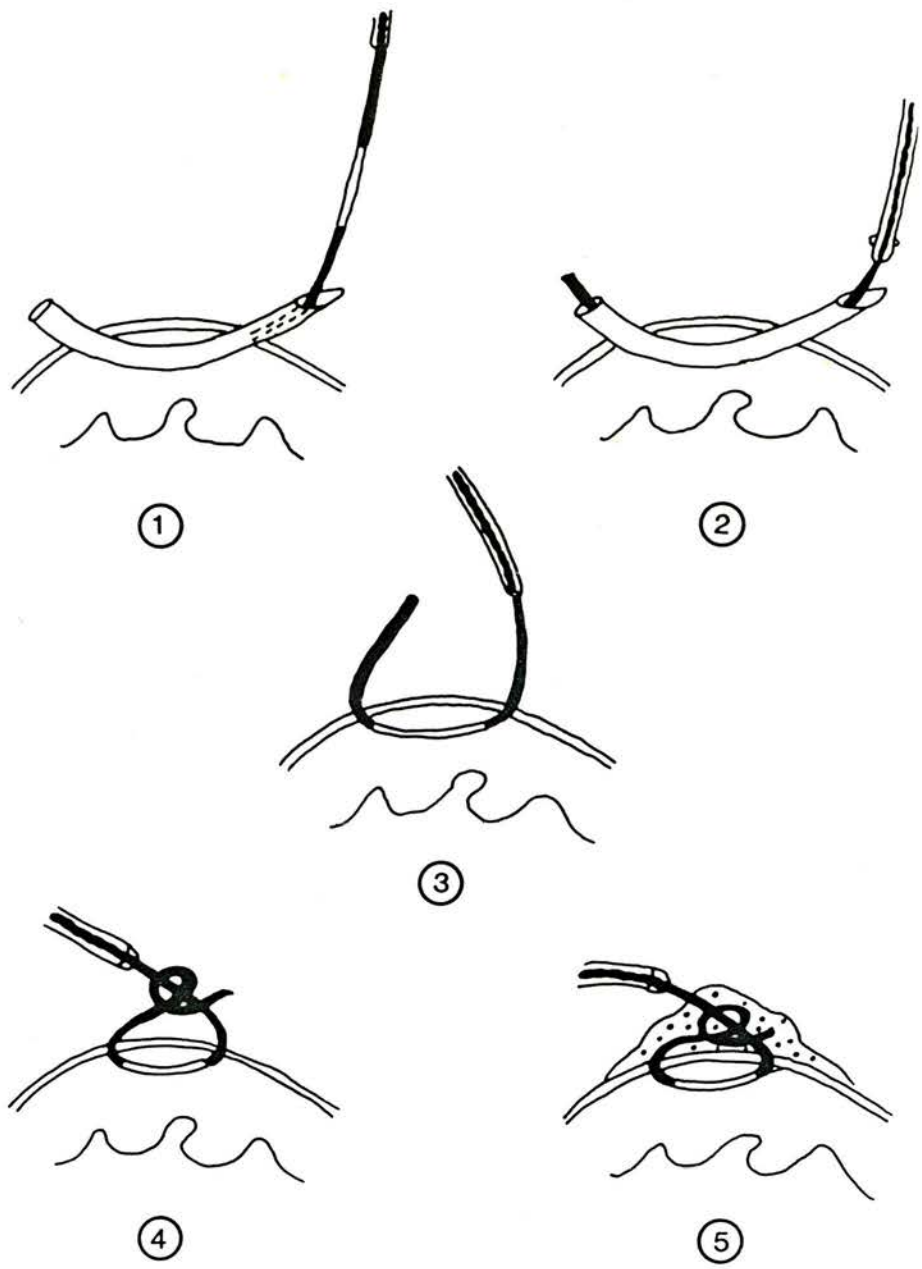
(2) The electrode was pushed until it emerged from the other end of the needle.

(3) The needle was taken out and the uninsulated part of the electrode was placed into the muscle tissue.

(4) The electrode was then knotted onto the serosa.

(5) After knotting, the implanted site was covered with M-coat D.

FIGURE 12.2
Technique of implantation of EMG electrodes.



implanted in a line in a group of three 1.0 cm apart (Ruckebusch, 1970). The electrodes were further insulated and stabilized with M-coat D. After implantation, a loop of the vinyl sheath of each set of electrodes was anchored within 5.0 cm of the implanting site by ligating the vinyl sheath to the wall of the implanted organ. This was performed to ensure the safety of the implanted site. The electrodes were then passed subcutaneously for a distance before exteriorizing through the skin at a suitable site preferably over the thoracic spine. After exteriorization, the vinyl sheath containing the electrodes was attached firmly to the skin in the vicinity of the point of emergence from the skin with superglue in order to keep the electrode position undisturbed. Each of the electrodes was connected with the plastic coated copper wire using pins since nichrome wire does not take ordinary solder. The free end of each copper wire was then connected to a junction box (25 way 'D' connectors, R.S. Components Ltd., Northants., England), which was securely tied into the fleece using the wool.

Surgical Approach to the Implanting Sites

Oesophagus: For implanting electrodes in the cervical oesophagus, the animal was positioned in dorsal recumbancy supported by two sand bags placed on either side of the body.

After preparing the site of operation, a longitudinal midline incision about 8.0 cm long was made. The subcutaneous tissue was divided by blunt dissection to expose the trachia. The oesophagus was then approached from the left side of the trachea by blunt dissection taking care to avoid injury to the important vessels and

nerves (e.g. jugular vein, carotid artery, branches of vagus nerves). A stomach tube was then introduced through the mouth into the oesophagus to ease its identification. A segment of the organ was dissected free. It was then lifted and held in position with two pairs of artery forceps placed underneath it at a distance of about 5.0 cm apart. The electrodes were then implanted following the technique described.

Gastro-oesophageal junction: To implant electrodes at this site, the animal was positioned in dorsal recumbancy and the operating table was adjusted so that the table was tilted about 20° towards the tail. A longitudinal midline incision of about 15-20 cm long was made extending from the xyphoid cartilage. The rumen was depressed manually and pushed caudally and held in place by an assistant. The gastro-oesophageal junction was identified by the presence of the stomach tube just posterior to the diaphragmatic hiatus. The electrodes were then sewn into the muscle as described. In one sheep, the electrodes were implanted at this site by a lateral approach through the left flank.

Rumen and reticulum: Sheep were positioned in right lateral recumbancy. The left flank was surgically prepared. A vertical incision of about 8-10 cm long was made in the skin and the rumen was exposed in the same way as described in connection with ruminal cannulation (Chapter 8). A pouch of rumen (dorsal sac) was exteriorized and held in position by applying tissue forceps and then the electrodes were implanted. The reticular electrodes were also implanted in a similar way.

The wounds were then closed using standard surgical techniques. The animal was kept in a separate pen for recovery from the effects of the anaesthetic. Once the animal recovered, it was transferred into either a metabolic cage or to a small individual pen to avoid excessive movement and possible interference to the implanted electrodes, especially by the other sheep. Recordings were made from two electrodes of each group of three with a Devices M19 multichannel recorder. The time constant was set at 0.1 sec, the sensitivity ranged from 500 μv - 2.5 mv and the high frequency cut at 10 Hz. Electromyograms were usually obtained two weeks after implantation and studied in both conscious and anaesthetized conditions.

Anaesthesia

Induction of anaesthesia was performed with halothane using a face mask and Magill system and maintained using a gaseous mixture of halothane, nitrous oxide and oxygen.

The mechanical activity of the implanted organs were sometimes recorded simultaneously with the EMG using catheter assembly 1 (Chapter 4). The experimental set up is shown in Figure 12.3. After a maximum of eight recordings, the sheep were killed and subjected to post-mortem examinations to verify the position of the implanted electrodes.

Results

Myoelectrical activity of the cervical oesophagus, gastro-oesophageal junction and reticulorumen were recorded from both conscious and anaesthetized sheep. The effects of the plane of anaesthesia on EMG discharge patterns from these organs were also investigated.

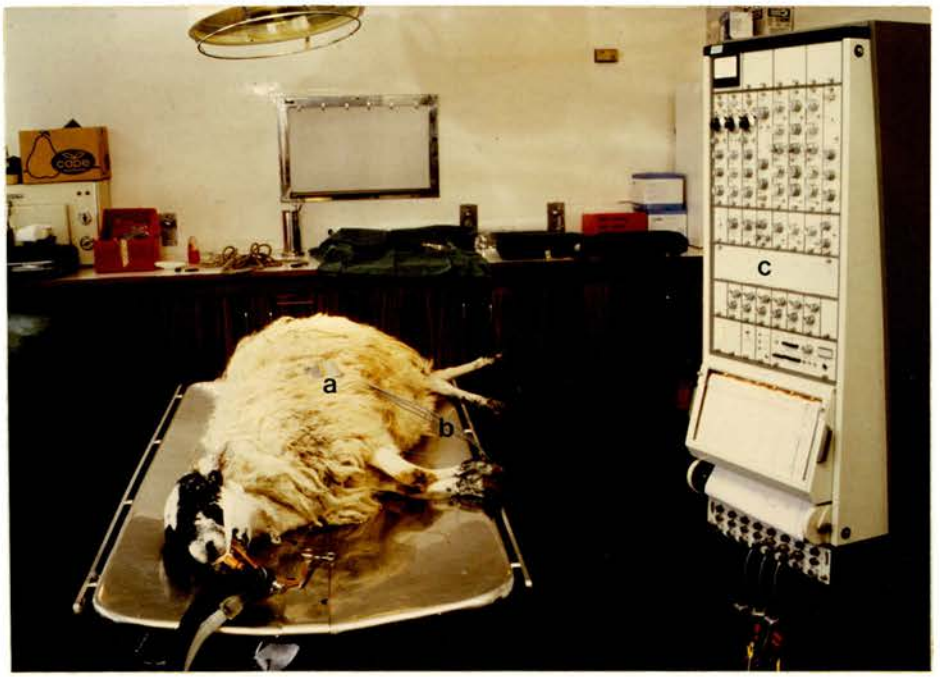
FIGURE 12.3

Experimental set up for electromyographic study.

a. Connecting box

b. Cables leading to the recorder from the connecting box.

c. Recorder


a
b
c

Electrical activities of the cervical oesophagus in conscious sheep and during light and deep planes of anaesthesia are illustrated in Figure 12.4. The oesophageal EMGs in conscious sheep were characterised by individual "spike discharges" (a spike is a graphical recording of transient variation in electric action potential characterised by a sharp peak). This pattern was also present in light anaesthesia while in deep anaesthesia, they were almost completely suppressed.

In two sheep, the electrodes were successfully positioned on the gastro-oesophageal junction as confirmed at necropsy. Simultaneous EMG recordings from the gastro-oesophageal junction, reticulum and rumen in the conscious sheep are presented in Figure 12.5. The EMG recordings from the junctional zone were characterised by a "continuous spike discharge" with a frequency of approximately 30 spikes per minute with occasional bursts of activity. The durations of these bursts were not consistent and ranged from 8 to 25 sec. These EMG bursts did not seem to be associated with either ruminal or reticular activities. The reticular spike discharge preceded those of ruminal ones. Occasionally, ruminal spike discharges were unrelated to reticular activities although the electrode positions were unchanged.

The effect of the plane of anaesthesia on the EMG activities of the gastro-oesophageal junction are shown in Figure 12.6. In light anaesthesia, the frequency and amplitude of spikes were greatly reduced when compared with the conscious state. These spikes continued to be present in deep anaesthesia.

FIGURE 12.4

Electromyographic activity of the cervical oesophagus in conscious sheep and during light and deep planes of anaesthesia (sensitivity - 500 μv; high frequency cut - 10 Hz; time constant - 0.1 sec). Single spikes were present in the conscious state. These oesophageal spikes were also present in light anaesthesia. In deep anaesthesia, the spiking activity of the cervical oesophagus was mostly abolished.

FIGURE 12.5

Electromyographic activities of the gastro-oesophageal junction, reticulum and rumen in conscious sheep (sensitivity - 1 mv; high frequency cut - 10 Hz; time constant - 0.1 sec). The junctional EMGs were characterized by continuous spiking activity (large negative spikes) with occasional bursts (bars). The frequency of individual spikes was approximately 30/minute and the duration of the burst varied from 8-25 sec. The reticular and ruminal spike bursts occurred at regular intervals, the reticular bursts preceded the ruminal ones. Some ruminal EMG bursts, however, were not associated with reticular bursts (arrows).

FIGURE 12.4

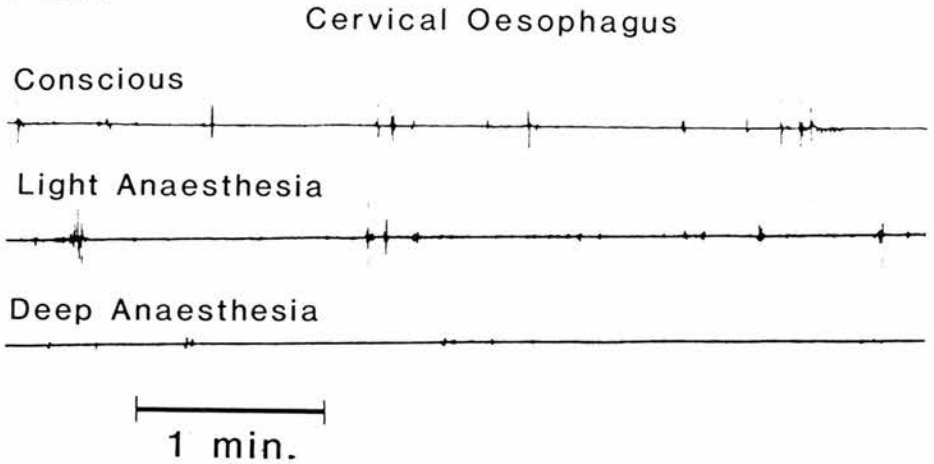


FIGURE 12.5

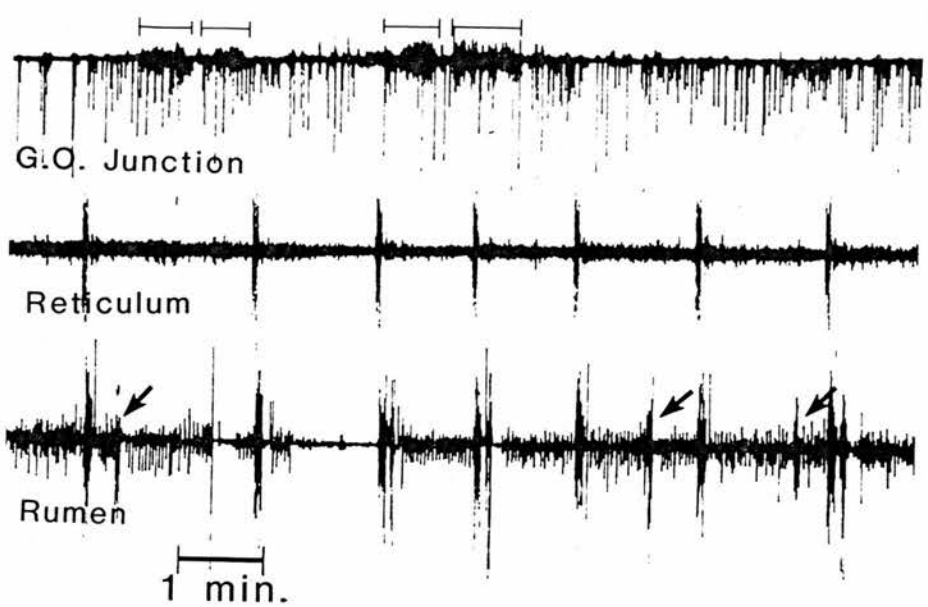


FIGURE 12.6

Electromyographic activities of the gastro-oesophageal junction (1.5 cm posterior to diaphragmatic hiatus) in the conscious sheep and during light and deep planes of anaesthesia (sensitivity - 1 mv; high frequency cut - 10 Hz; time constant - 0.1 sec). The continuous spiking activity in light anaesthesia was reduced in both frequency and amplitude and remained during deep anaesthesia.

FIGURE 12.7

Electromyographic activities of the reticulum in the conscious sheep and during light and deep planes of anaesthesia (sensitivity - 1 mv; high frequency cut - 10 Hz; time constant - 0.1 sec). The regular reticular bursts were reduced in frequency in light anaesthesia while in deep anaesthesia, these activities were totally abolished.

FIGURE 12.6

Gastro-oesophageal Junction

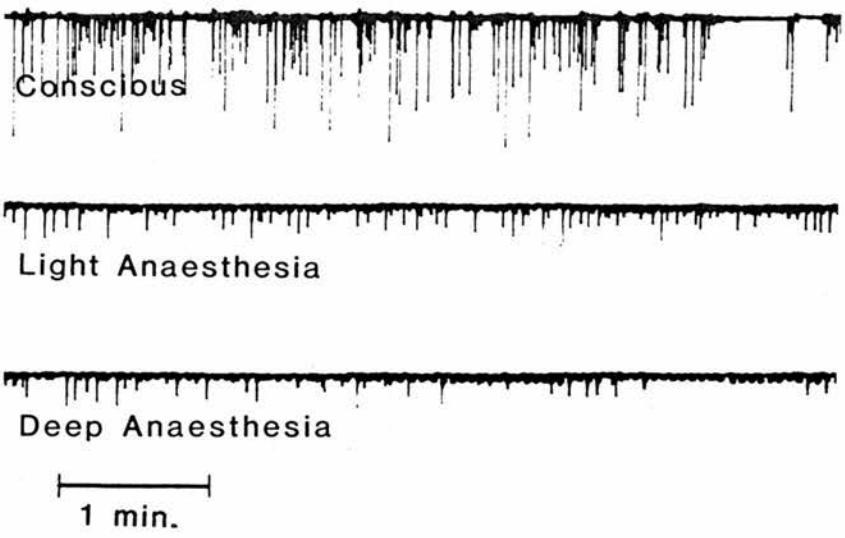
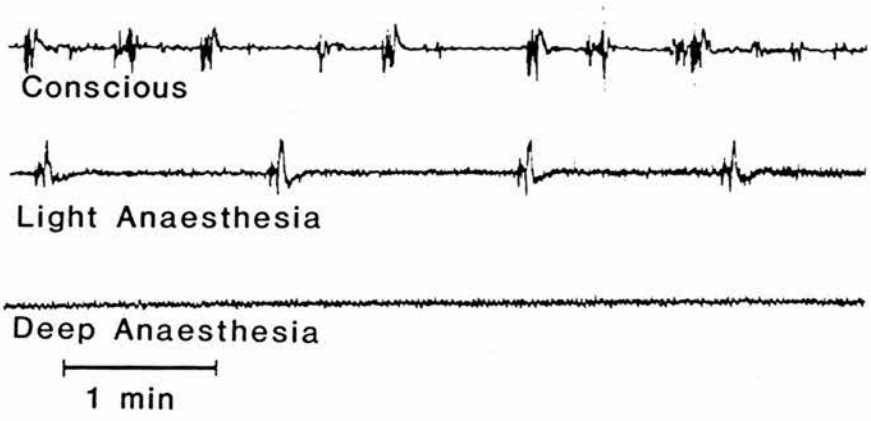


FIGURE 12.7

Reticulum



In two sheep, the electrodes thought to be implanted at the gastro-oesophageal junctional area were actually found to be in the anterior reticulum at necropsy. The EMGs from the reticulum in conscious sheep and during light and deep planes of anaesthesia are presented in Figure 12.7. These activities were different from those of gastro-oesophageal junction (Figure 12.6) and were characterized by "bursts" of spike discharge. The rate of this discharge was greatly reduced in light anaesthesia while in deep anaesthesia they were totally abolished.

The electrical activities of the rumen in conscious sheep and during light and deep anaesthesia are presented in Figure 12.8. The spike discharge associated with rumen contraction was reduced in both rate and amplitude in light anaesthesia as compared with the conscious state. In deep anaesthesia, the ruminal electrical activity was abolished.

Simultaneous recordings of electrical and mechanical activities of the rumen and gastro-oesophageal junction were performed and these are presented in Figure 12.9. The ruminal pressure waves were preceded by a burst of spike discharge. The LOS pressure waves, however, were not associated with any burst of activity in the gastro-oesophageal junction although these pressure waves were associated with ruminal pressure waves.

Summary

In summary, these results demonstrate that in conscious sheep, the cervical oesophagus usually produced single EMG spikes and no spike bursts were observed. In light anaesthesia, the spiking

FIGURE 12.8

Electrical activities of the rumen in the conscious sheep and during light and deep planes of anaesthesia (sensitivity - 1 mv; high frequency cut - 10 Hz; time constant - 0.1 sec). The ruminal spike bursts in light anaesthesia were reduced in frequency and amplitude as compared with the conscious state, while in deep anaesthesia these spike bursts were totally abolished.

FIGURE 12.9

Simultaneous electrical and mechanical records from the rumen and lower oesophageal sphincter in light anaesthesia (sensitivity - 1 mv; high frequency cut - 10 Hz; time constant - 0.1 sec). The EMGs of the gastro-oesophageal junction 1.5 cm posterior to diaphragmatic hiatus were compared with the pressure changes of the LOS in these tracings. The ruminal pressure waves were always preceded by ruminal spike bursts in light anaesthesia. Some weak spikes, however, were present apart from ruminal pressure waves. The mechanical activities of the LOS were not associated with any EMG burst of the gastro-oesophageal junction. The junctional zone showed some individual spikes which were not related to LOS pressure waves. The LOS pressure waves, however, were always associated with the ruminal pressure waves.

FIGURE 12.8

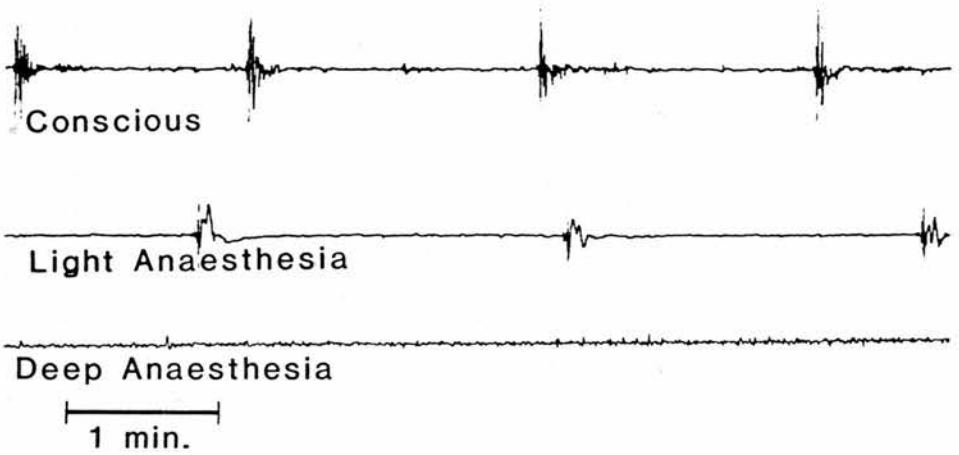
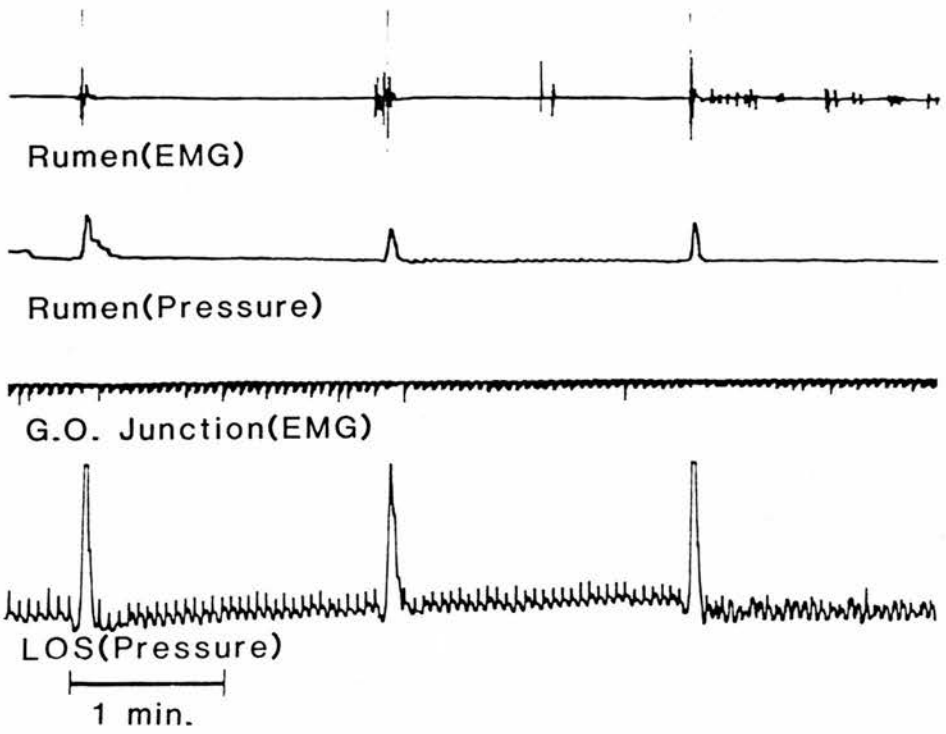


FIGURE 12.9



activities were slightly reduced and were almost abolished in deep anaesthesia. The measurement of thoracic oesophageal pressure waves were not found to be associated with these EMG changes in light anaesthesia.

Ruminal and reticular EMGs in the conscious sheep were characterized by the presence of bursts of spike discharges at regular intervals. The reticular spike bursts were usually found to precede the ruminal ones. Occasionally, the ruminal bursts occurred apart from reticular bursts. These reticuloruminal EMGs were also present in light anaesthesia although the frequency and the amplitudes were reduced. Simultaneous manometry and EMGs of the rumen showed consistent correlation i.e. ruminal pressure waves were always associated with EMG discharge. In deep anaesthesia, however, these pressure waves and the EMG bursts were totally abolished.

Electromyographic activity of the gastro-oesophageal junction from the conscious animal was usually characterized by continuous spiking activity with occasional bursts of EMG discharge. This pattern was completely different from those of the rumen and reticulum. In light anaesthesia, the spiking activity was present but the frequency and the amplitude of these spikes were reduced. These activities were also present in deep anaesthesia. There was no association between the spiking activity of the junctional area and the pressure changes in either the lower oesophageal sphincter or in the rumen. These activities were also not associated with breathing.

Discussion

This investigation supports some of the results obtained in the manometric studies of the activities of the oesophagus and reticulo-rumen (Chapters 9 and 10). The demonstration of the presence of oesophageal and ruminal motility in light anaesthesia and their abolition in deep anaesthesia have been consistently supported by the EMG studies.

In the present study, the electrical activities were associated with mechanical activities which are in agreement with the findings of Grivel and Ruckebusch (1972); Bueno, Ferre, Ruckebusch, Genton and Pascaud (1981) in conscious sheep. Rattan, Gidda and Goyal (1983) however, reported that electrical and mechanical activity may not necessarily occur in the same group of muscle cells. These workers stated that depolarization is usually associated with spike burst but may occur without any spike burst or contraction.

Itabisashi (1970) suggested that the absence of myoelectrical activity does not always exclude the occurrence of mechanical contraction. He stated that when the reticulum wall was moved up and down by hand, no burst was recorded but only slow deflections. The bursts, therefore, cannot be regarded as the movement artifact.

The initiation of electrical spike discharges is uncertain. Bortoff (1975) suggested that the rhythmical nature of the gastro-intestinal contractions is due to the slow wave periodically increasing and decreasing the excitability of the muscle membrane and that if the excitability is sufficiently increased, spike potentials are generated and a contraction ensues.

The oesophageal EMGs in conscious and light anaesthesia were usually single spikes. These spikes in conscious sheep were mostly due to swallowing and eructation. As the eructation reflex is inhibited in anaesthesia, the presence of spikes in light anaesthesia is either due to swallowing or to local muscular twitching.

The thoracic oesophageal pressure waves in light anaesthesia were not usually associated with cervical oesophageal spike discharge. It has been demonstrated in Chapter 9 that the oesophageal pressure waves were mostly present in the thoracic oesophagus which may explain why the EMGs were not associated with pressure waves.

The frequency and amplitude of rumen and reticulum contractions were greatly reduced in light anaesthesia as compared with the conscious state. This may be due to suppression of the motor centres in the brain by halothane.

The EMGs of the gastro-oesophageal junction were completely different from those of reticulorumen and cervical oesophagus. Similar recordings (continuous spiking activity) have been reported in the opossum LOS by Asoh and Goyal (1978).

In the present investigation, the pressure changes in the LOS were not associated with any EMG activity of the gastro-oesophageal junction. The possible explanations are (i) it may be the characteristic behaviour of the gastro-oesophageal junction or (ii) the electrodes implanted at this site were not at the high pressure zone, in which case the electrical activities recorded might belong to diaphragmatic hiatus. Hellemans et al. (1968) reported that relaxation of LOS is not associated with any electrical change in the monogastric animals.

The implantation of the electrodes at the gastro-oesophageal junction was relatively difficult in comparison with other implanting sites. Although starved sheep were used, the reticulorumen still concealed the site of implantation. Tilting of the operating table towards the tail was helpful. At necropsy, the electrodes implanted through midline approach were found at the appropriate site but those implanted by lateral approach through the left flank were found on the reticulum about 3.0 cm posterior to the junction.

The electrodes implanted at the gastro-oesophageal junction were superficial and included external muscle only as observed at necropsy. Therefore, the recordings of this site were probably not the exact representation of LOS. Further studies are essential to specifically characterize the electromyographic activity of the gastro-oesophageal junction.

Conclusions

(i) Electromyographic activity of the oesophagus and reticulorumen persists in light anaesthesia but is abolished when the anaesthesia is deepened.

(ii) Electrical activities of the gastro-oesophageal junction mostly comprise of continuous spiking activity regardless of reticuloruminal contractions or plane of anaesthesia.

(iii) Ruminal EMG is indicative of mechanical activity.

(iv) Electromyography does not appear to be a suitable technique in monitoring the different patterns of activities of the above organs especially because no EMG change is seen during muscle relaxation.

CHAPTER THIRTEEN

THE EFFECTS OF INTRAVENOUS ANAESTHETIC AGENTS ON GASTRO-OESOPHAGEAL REFLUX

Introduction

In ruminants, intravenous anaesthetics have been almost replaced by inhalation anaesthetics at the present time. Induction of anaesthesia, however, is still mainly performed by the administration of intravenous anaesthetic agents. These intravenous agents are also extensively used in the Third World countries as the sole anaesthetic agents. Pentobarbitone, thiopentone, chloral hydrate and more recently alphaxalone/alphadolone are the drugs commonly used. Pentobarbitone and thiopentone are used in sheep for short lasting surgical interventions. Their use in ruminant anaesthesia has been threatened by some observational impressions that intravenous barbiturates tend to provoke gastro-oesophageal reflux (Leek, 1975). There is no experimental evidence to suggest that barbiturates, chloral hydrate or alphaxalone/alphadolone stimulate reflux of rumen contents into the oesophagus or influence the activities of the oesophagus and reticulorumen. Experiments were performed to study the influence of a selection of these anaesthetics on the occurrence of gastro-oesophageal reflux in the anaesthetized sheep.

Materials and Methods

Ten sheep were anaesthetized on a total of 40 occasions in the present study. All these animals were housed in a similar fashion as described in the general methods (Chapter 4). None were starved or given any premedication prior to anaesthesia.

Anaesthesia

Animals were physically restrained on the operating table in right lateral recumbancy. Once the animal was relatively settled,

the area about the recurrent tarsal vein was clipped and sprayed with chlorehexidine gluconate (Hibitane , ICI, Great Britain). The leg above the clipped area was compressed by an assistant to raise the recurrent tarsal vein. A 16-gauge catheter (Vygon Intranule Laboratoires Pharmaceutiques, Ecouen, France) was introduced into the vein and was secured with a thread fastened around the limb. The administration of the intravenous anaesthetic was made through the cannula. The doses of the different anaesthetics used are given below:

Pentobarbitone sodium (Nembutal, Ceva Ltd., England)
Initial dose: 30 $mg.kg^{-1}$

Thiopentone sodium (Intraval Sodium, May and Baker, Dagenham, England)
Initial dose: 20 $mg.kg^{-1}$ (2.5% solution)

Chloral hydrate - Magnesium sulphate
Initial dose for chloral hydrate: 100 $mg.kg^{-1}$ (12% solution)
Initial dose for $MgSO_4$: 100 $mg.kg^{-1}$ (6% solution)

Alphaxalone/alphadolone (Saffan, Glaxovet Ltd.)
Initial dose: 3-6 $mg.kg^{-1}$

After administration of the initial dose of the anaesthetic, the larynx was sprayed with Xylocaine spray (Astra Chemicals Ltd., Watford, Herts., England) to cause anaesthesia of the glottis which prevents spasm. The trachea was then intubated with a cuffed endotracheal tube to ensure the patency of the airway in the same technique as described in the general methods (Chapter 4). Thus the animal was able to utilize room air for breathing. The additional doses of the drugs were administered when necessary to prolong anaesthesia up to 60 minutes.

The intraluminal pressures of the oesophagus and rumen were measured throughout anaesthesia using catheter assembly 2 as described in the general methods (Chapter 4).

The data obtained were analysed by using the Student's t-test.

Results

Pentobarbitone: Intraruminal pressure built up during pentobarbitone anaesthesia over a period of 60 minutes is presented in Table 13.1. The pressure increased to 11.2 ± 1.7 (SD) mmHg at 60 minutes from an initial level of 8.5 ± 1.3 (SD) mmHg (an increase of 2.7 mmHg).

Out of ten anaesthetics, reflux occurred in six cases (60%). Maximum pressure gradient between rumen and thoracic oesophagus in each anaesthetic was calculated. Reflux was associated with maximum pressure gradient in 60% of anaesthetics (Table 13.2).

The motor activities of the oesophagus were completely abolished at all levels of anaesthesia. The amplitude and frequency of ruminal pressure waves were also greatly suppressed. In two anaesthetics, extremely weak pressure waves were found to occur in light anaesthesia (Figure 13.1). In light anaesthesia, stimulated deglutition by external pharyngeal palpation did not produce any peristaltic waves. No oesophageal pressure waves were observed associated with reflux in any depth of anaesthesia. Occasionally, ruminal pressure waves were found associated with reflux (Figure 13.2) and the oesophageal tracing only recorded the intrapleural pressure changes associated with respiration.

Table 13.1 Intraruminal pressure built up during pentobarbitone anaesthesia over a period of 60 minutes

Sheep No.	Initial pressure (mmHg)	Pressure after 10 mins (mmHg)	Pressure after 20 mins (mmHg)	Pressure after 30 mins (mmHg)	Pressure after 40 mins (mmHg)	Pressure after 50 mins (mmHg)	Pressure after 60 mins (mmHg)
604E	9.0	9.5	10.0	10.5	10.5	10.5	13.5
502E	7.5	8.0	9.0	9.5	9.5	10.0	10.5
505E	9.0	9.5	11.0	11.0	11.5	11.5	12.5
509E	10.0	9.5	10.0	10.5	10.5	10.5	11.0
530E	9.5	10.0	10.5	11.0	12.5	12.5	13.0
498E	5.5	6.5	7.0	7.0	7.5	7.5	7.5
536E	9.5	9.5	9.5	10.0	10.0	12.0	12.0
528E	9.0	9.5	10.5	10.5	11.0	11.0	11.0
510E	7.5	8.5	8.5	9.5	10.0	10.0	10.0
506E	8.5	8.5	9.0	9.0	9.5	10.5	10.5
Mean	8.5	8.9	9.5	9.85	10.25	10.6	11.15
S.D.	1.33	1.04	1.17	1.20	1.33	1.37	1.73

Table 13.2 Maximum pressure gradient and occurrence of reflux during pentobarbitone anaesthesia

Sheep No.	Maximum Pressure (mmHg)			Occurrence of reflux
	Rumen	Thoracic oesophagus	Rumeno-oesophageal gradient	
604E	17.0	0.5	16.5	+
502E	9.5	-0.5	10.0	+
505E	10.0	0.0	10.0	+
509E	11.0	-2.5	13.5	-
530E	9.5	-2.5	12.0	+
498E	7.0	-2.5	9.5	+
536E	9.5	0.0	9.5	+
528E	11.0	0.0	11.0	-
501E	7.5	-0.5	8.0	-
506E	8.5	-0.5	9.0	-
Mean	10.05	-0.85	10.9	+ 60%
S.D.	2.77	1.17	2.51	- 40%

FIGURE 13.1

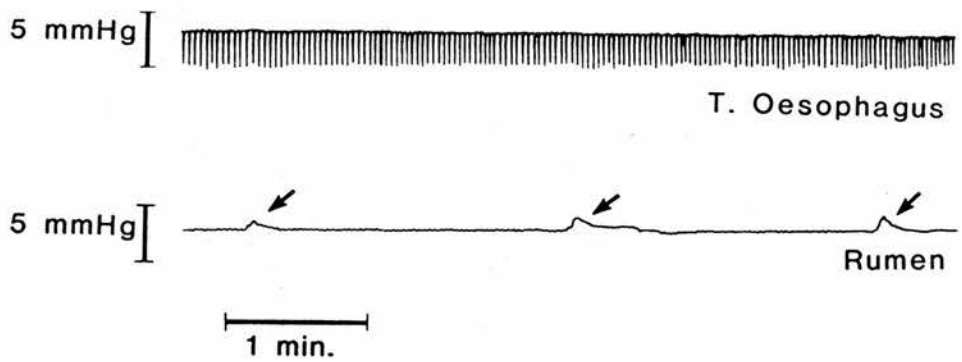
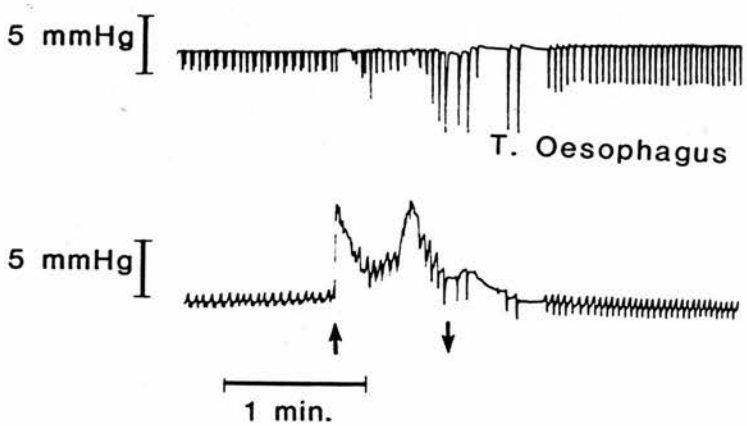


FIGURE 13.2



Thiopentone: Intraruminal pressure built up during thiopentone anaesthesia is presented in Table 13.3. The pressure increased to 11.9 ± 1.5 (SD) mmHg at 60 minutes from an initial level of 10.0 ± 1.4 (SD) mmHg (an increase of 1.9 mmHg).

Out of ten anaesthetics in five sheep, reflux occurred in seven cases (70%). Maximum pressure gradient between rumen and thoracic oesophagus and the occurrence of reflux were recorded in each anaesthetic (Table 13.4). Reflux was associated with maximum pressure gradient in 40% of anaesthetics.

Like pentobarbitone, thiopentone also caused total abolition of oesophageal activity. Ruminal pressure waves were totally abolished in both light and deep levels of anaesthesia. The thoracic oesophageal pressures always showed reduction in pressure associated with inspiration. At times, these changes increased in amplitude (Figure 13.3). The pressure changes of the rumen and thoracic oesophagus associated with reflux are shown in Figure 13.4. There were no associated pressure waves. The tone of both the oesophagus and rumen, however, was slightly increased. In 10% of the anaesthetics, the occurrence of reflux was coincided with the cessation of breathing (Figure 13.5).

Chloral hydrate-Magnesium sulphate: Intraruminal pressure built up during chloral hydrate-$MgSO_4$ anaesthesia is presented in Table 13.5. This pressure increased to 11.7 ± 1.5 (SD) mmHg from an initial level of 9.8 ± 1.2 (SD) mmHg (an increase of 1.9 mmHg).

Reflux occurred in six anaesthetics out of ten (60%). Maximum pressure gradient and the occurrence of reflux in each anaesthetic

Table 13.3 Intraruminal pressure built up during thiopentone anaesthesia over a period of 60 minutes

Sheep No.	Initial pressure (mmHg) n = 2	Pressure after 10 mins (mmHg) n = 2	Pressure after 20 mins (mmHg) n = 2	Pressure after 30 mins (mmHg) n = 2	Pressure after 40 mins (mmHg) n = 2	Pressure after 50 mins (mmHg) n = 2	Pressure after 60 mins (mmHg) n = 2
616E	9.0	9.5	9.75	9.75	9.75	10.0	10.5
545E	12.25	13.25	13.25	13.75	13.75	14.0	14.25
551E	9.5	10.5	11.0	11.5	11.75	11.75	12.25
514E	10.25	10.5	11.25	11.5	11.5	11.75	11.75
546E	9.0	9.5	9.75	10.25	10.5	10.5	10.5
Mean	10.0	10.65	11.0	11.35	11.45	11.6	11.85
S.D.	1.35	1.53	1.43	1.54	1.51	1.54	1.54

Table 13.4 Maximum pressure gradient and the occurrence of reflux during thiopentone anaesthesia

Anaesthetic No.	Maximum Pressures (mmHg)			Occurrence of reflux
	Rumen	Thoracic oesophagus	Rumeno-oesophageal gradient	
1	10.5	0.0	10.5	-
2	9.5	-1.0	10.5	-
3	15.0	1.0	14.0	+
4	13.0	0.0	13.0	+
5	11.5	-1.0	12.5	-
6	11.5	0.5	11.0	+
7	13.0	0.5	12.5	-
8	10.0	0.0	10.0	+
9	8.0	-0.5	8.5	-
10	12.5	0.0	12.5	-
Mean	11.45	-0.05	11.5	+ 40%
S.D.	2.03	0.64	1.66	- 60%

Table 13.5 Intraruminal pressure built up during chloral hydrate-magnesium sulphate anaesthesia

Sheep No.	Initial pressure (mmHg) n = 2	Pressure after 10 mins (mmHg) n = 2	Pressure after 20 mins (mmHg) n = 2	Pressure after 30 mins (mmHg) n = 2	Pressure after 40 mins (mmHg) n = 2	Pressure after 50 mins (mmHg) n = 2	Pressure after 60 mins (mmHg) n = 2
498E	9.0	9.0	9.25	9.5	10.0	10.25	10.25
111E	10.25	10.75	11.0	11.75	11.75	12.25	12.25
509E	11.75	12.5	13.0	13.5	14.25	14.0	14.0
528E	9.0	9.25	9.5	9.75	10.25	10.25	10.75
506E	9.0	10.5	10.75	11.25	11.25	11.25	11.25
Mean	9.8	10.4	10.7	11.15	11.5	11.6	11.7
S.D.	1.21	1.39	1.49	1.62	1.69	1.57	1.48

FIGURE 13.3

Thoracic oesophageal and ruminal pressure changes during light thiopentone anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). Neither thoracic oesophagus nor rumen showed any pressure waves. The thoracic oesophagus, however, showed intermittent decrease in pressure associated with deep inspirations (arrows).

FIGURE 13.4

Thoracic oesophageal and ruminal pressure changes during thiopentone anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows show the timing of brief periods of reflux. There were no additional thoracic oesophageal or ruminal pressure waves at the point of reflux. At the second arrow, however, there was a little increase in tone of the thoracic oesophagus. The corresponding ruminal tracing showed a little relaxation prior to reflux followed by a slight increase in tone.

FIGURE 13.5

Thoracic oesophageal and ruminal pressure changes during thiopentone anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows show the occurrence of reflux. Note the cessation of breathing associated with reflux.

FIGURE 13.3

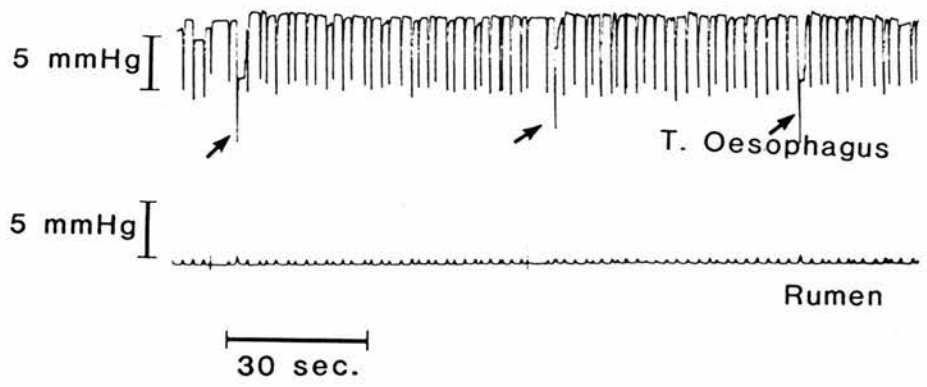


FIGURE 13.4

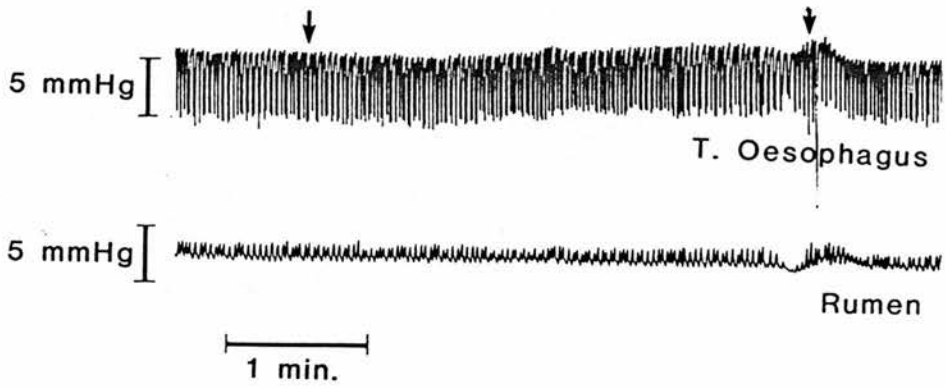
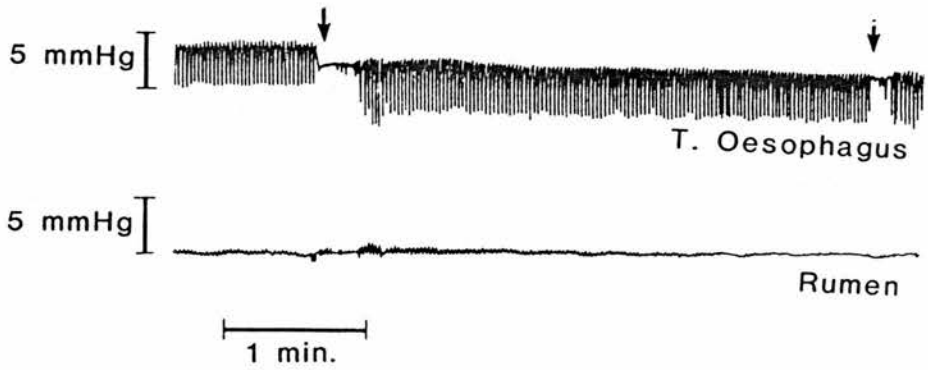


FIGURE 13.5



are presented in Table 13.6. Reflux occurred in association with maximum pressure gradient in 40% of cases.

During light chloral hydrate anaesthesia, oesophageal and ruminal pressure waves were frequently present (Figure 13.6). In deep anaesthesia, both the oesophageal and ruminal pressure waves were abolished. As the anaesthesia lightened, ruminal pressure waves reappeared with gradual increase in amplitude (Figure 13.7). During light anaesthesia when additional small doses of chloral hydrate were administered, the ruminal pressure waves were instantly abolished and the duration of abolition depended on the amount of drug administered (Figure 13.8).

The pressure changes of thoracic oesophagus and rumen associated with reflux are shown in Figure 13.9. The tone of the thoracic oesophagus was reduced at reflux. The thoracic oesophageal relaxation, however, was usually absent at reflux. In 10% of cases, reflux occurred following each ruminal pressure wave (Figure 13.10).

The direction of the thoracic oesophageal pressure waves during light chloral hydrate anaesthesia was invariably peristaltic (Figure 13.11).

Alphaxalone/alphadolone (Saffan): Intraruminal pressure built up during these anaesthetics is presented in Table 13.7. The pressure increased to 12.2 ± 0.9 (SD) mmHg from an initial level of 8.7 ± 0.8 (SD) mmHg (an increase of 3.5 mmHg).

Out of ten anaesthetics, reflux occurred in four cases (40%). Maximum pressure gradients and the occurrence of reflux are presented in Table 13.8. Thirty percent of the reflux was associated with maximum pressure gradients.

Table 13.6 Maximum pressure gradient and the occurrence of reflux during chloral hydrate anaesthesia

Anaesthetic No.	Maximum Pressures (mmHg)			Occurrence of reflux
	Rumen	Thoracic oesophagus	Rumeno-oesophageal gradient	
1	10.5	-1.0	11.5	+
2	10.5	0.0	10.5	-
3	11.5	-1.0	12.5	+
4	13.0	-1.0	14.0	-
5	15.0	0.0	15.0	+
6	12.5	0.5	12.0	+
7	8.0	-1.0	9.0	-
8	13.0	0.0	13.0	-
9	10.0	1.0	11.0	-
10	12.5	0.0	12.5	-
Mean	11.65	-0.25	12.1	+ 40%
S.D.	1.97	0.71	1.72	- 60%

Table 13.7 Intraruminal pressure built up during alphaxalone/alphadolone anaesthesia

Sheep No.	Initial pressure (mmHg) n = 2	Pressure after 10 mins (mmHg) n = 2	Pressure after 20 mins (mmHg) n = 2	Pressure after 30 mins (mmHg) n = 2	Pressure after 40 mins (mmHg) n = 2	Pressure after 50 mins (mmHg) n = 2	Pressure after 60 mins (mmHg) n =2
506E	8.75	9.25	10.0	10.5	11.25	11.25	12.75
498E	8.0	8.25	8.75	9.25	10.25	10.5	11.25
604E	8.0	8.75	10.25	10.0	10.0	11.5	11.5
505E	10.0	10.75	11.5	11.75	11.75	13.0	13.5
528E	8.5	10.0	10.25	11.0	11.5	11.75	12.0
Mean	8.65	9.4	10.15	10.5	10.95	11.6	12.2
S.D.	0.82	0.99	0.97	0.95	0.77	0.91	0.92

Table 13.8 Maximum pressure gradient and the occurrence of reflux during alphaxalone/alphadolone anaesthesia

Anaesthetic No.	Maximum Pressures (mmHg)			Occurrence of reflux
	Rumen	Thoracic oesophagus	Rumeno-oesophageal gradient	
1	12.5	0.0	12.5	-
2	12.5	1.0	11.5	-
3	12.0	-1.0	13.0	-
4	8.0	-3.5	11.5	+
5	8.0	0.0	8.0	-
6	14.5	1.0	13.5	-
7	15.0	-1.0	16.0	+
8	11.5	-0.5	12.0	+
9	11.0	-4.5	15.5	-
10	12.5	0.0	12.5	-
Mean	11.75	-0.85	12.6	+ 30%
S.D.	2.32	1.81	2.23	- 70%

FIGURE 13.6

Thoracic oesophageal and ruminal pressure changes during light chloral hydrate-$MgSo_4$ anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The thoracic oesophageal and ruminal pressure waves were frequently present.

FIGURE 13.7

Thoracic oesophageal and ruminal pressure changes as anaesthesia lightened. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). With the lightening of anaesthesia, the ruminal pressure waves reappeared with a gradual increase in amplitude. The thoracic oesophagus in this section of trace was not active.

FIGURE 13.8

Thoracic oesophageal and ruminal pressure changes during light chloral hydrate-$MgSo_4$ anaesthesia associated with fresh injections of the anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows show the timing of fresh administration of the anaesthetic (3 mls at the first arrow and 5 mls at the second arrow). Note that the ruminal pressure waves were abolished completely following the fresh injections. The duration of abolition of ruminal pressure waves depended on the amount of the drug administered.

FIGURE 13.6

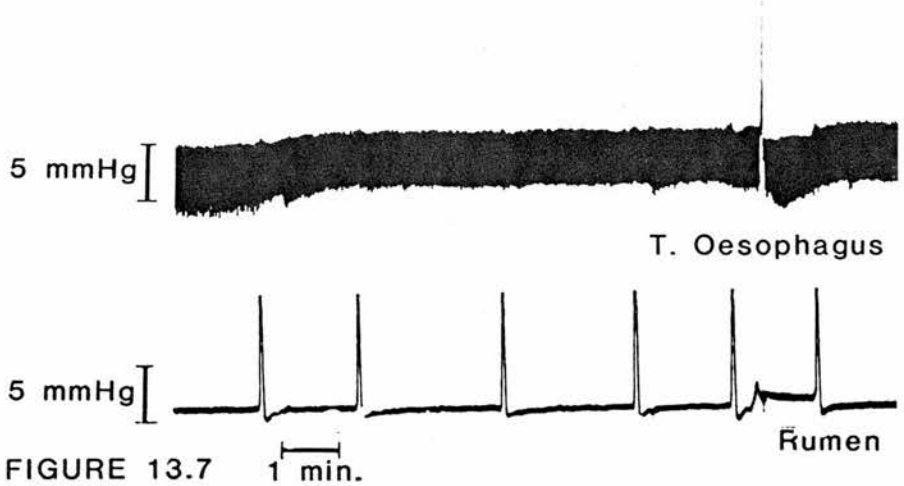


FIGURE 13.7

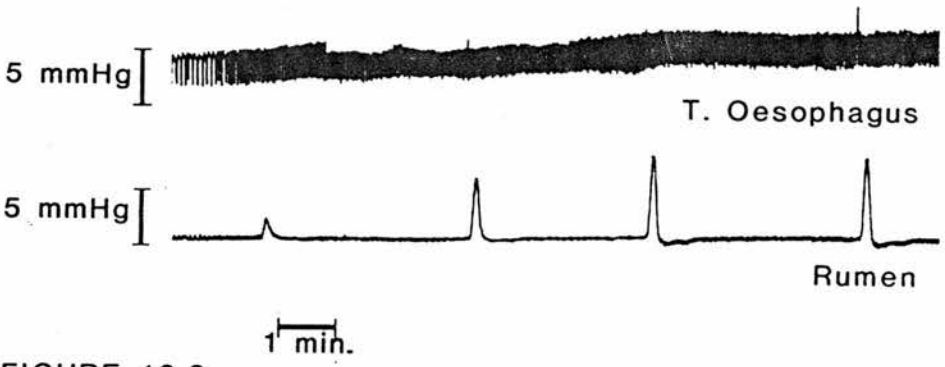


FIGURE 13.8

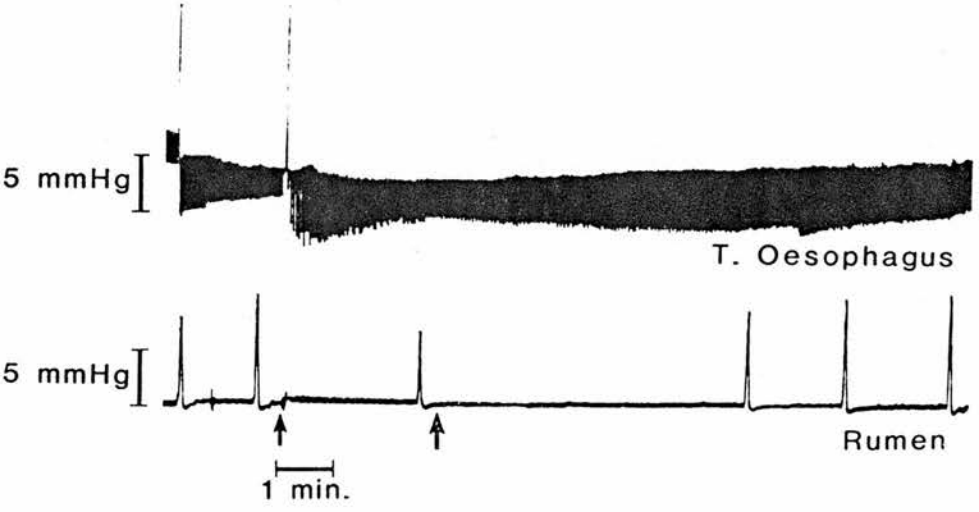


FIGURE 13.9

Thoracic oesophageal and ruminal pressure changes associated with reflux during chloral hydrate-$MgSo_4$ anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the onset and termination of reflux. The tone of thoracic oesophagus was reduced during the period of reflux. The ruminal tracing did not show any associated changes.

FIGURE 13.10

Thoracic oesophageal and ruminal pressure changes associated with reflux in light chloral hydrate-$MgSo_4$ anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows show the timing of brief refluxes. Reflux occurred consistently after every ruminal pressure wave. The thoracic oesophagus was quiescent during these refluxes.

FIGURE 13.11

Direction of thoracic oesophageal pressure waves during chloral hydrate-$MgSo_4$ anaesthesia. Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves in the anterior thoracic oesophagus preceded those of posterior thoracic oesophagus, i.e. the waves were peristaltic.

FIGURE 13.9

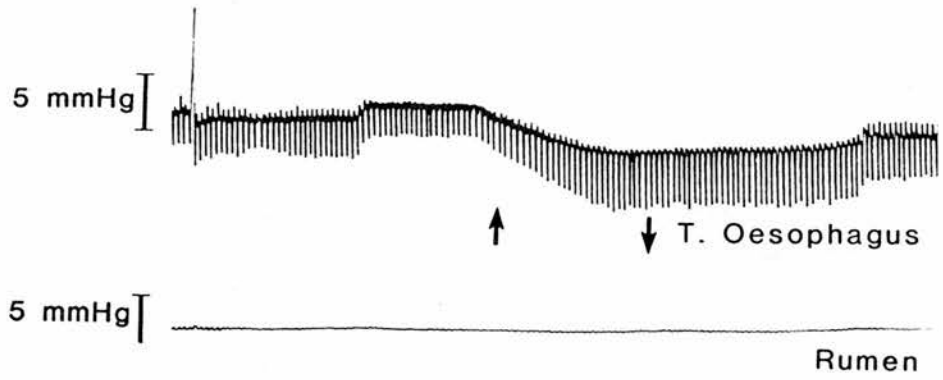


FIGURE 13.10

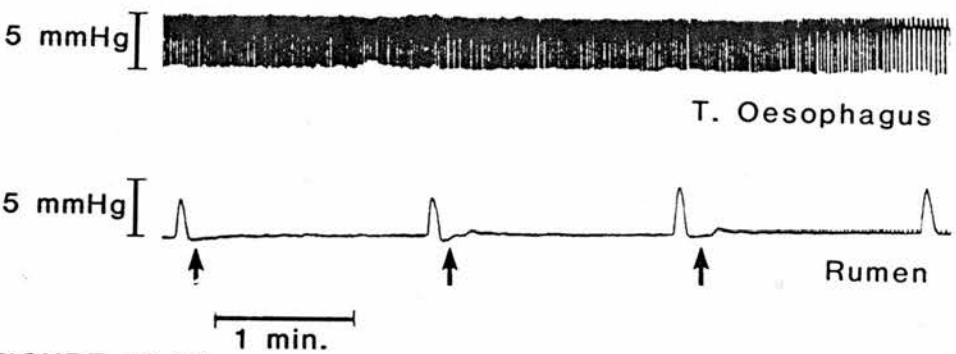
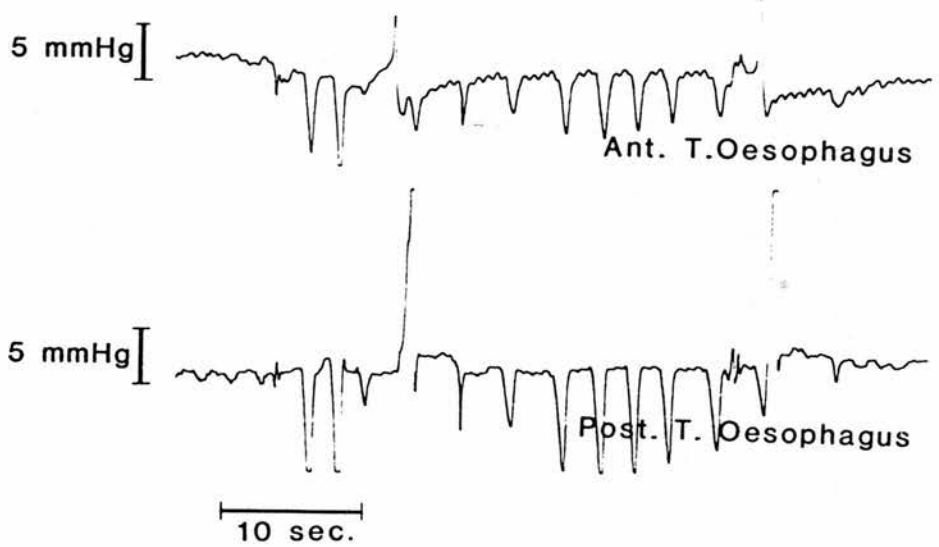


FIGURE 13.11



The oesophageal and ruminal pressure waves were present during light alphaxalone/alphadolone anaesthesia. These oesophageal pressure waves were frequently associated with reflux in light anaesthesia (Figure 13.12). In about 15% of cases, reflux was associated with deep inspirations (Figure 13.13). The direction of oesophageal pressure waves was always peristaltic (Figure 13.14) and the velocity of these waves was 24.0 cm/sec. The ruminal pressure waves during this anaesthetic, however, were infrequent.

The relationship of the intraruminal pressures which were built up during pentobarbitone, thiopentone, chloral hydrate-$MgSO_4$ and alphaxalone/alphadolone anaesthesia is presented in Figure 13.15. The largest intraruminal pressure build up was found in alphaxalone/alphadolone anaesthesia and the smallest with thiopentone anaesthesia. This difference was statistically not significant.

The influence of intravenous anaesthetic agents on reflux and gastro-oesophageal motor activities are summarised in Table 13.9.

Discussion

Pentobarbitone and thiopentone: Barbiturates appear to be a good short acting intravenous anaesthetic agent in sheep providing the patency of the airway is maintained with a cuffed endotracheal tube. The short duration (about 25 minutes) of pentobarbitone anaesthesia in sheep is due to the rapid metabolism of the drug in the liver (Rae, 1962). In the present study, reflux occurred in 60% of cases in the pentobarbitone group in contrast to only 6.6% reported by Harrison (1964) in pentobarbitone anaesthetized sheep. This significantly lower incidence of reflux in Harrison's study may be due

Table 13.9 Influence of intravenous anaesthetic agents on reflux and gastro-oesophageal motor activities

Anaesthetic agents	Number of anaesthetics	Number of anaesthetics with reflux	Percentage of occurrence	% of reflux associated with maximum pressure gradient	Oesophageal motility using open tip catheter (OTC) + Present - Absent	Ruminal motility (OTC) + Present - Absent
Pentobarbitone	10	6	60	60	-	+
Thiopentone	10	7	70	40	-	-
Chloral hydrate + $MgSO_4$	10	6	60	40	+	+
Alphaxalone/ alphadolone	10	4	40	30	+	+

FIGURE 13.12

Thoracic oesophageal and ruminal pressure changes associated with reflux during light alphaxalone/alphadolone anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The arrows indicate the onset and termination of reflux. The thoracic oesophageal pressure waves commenced before the onset of reflux and persisted during the period of reflux and continued after the termination of reflux.

FIGURE 13.13

Thoracic oesophageal and ruminal pressure changes associated with reflux during light alphaxalone/alphadolone anaesthesia. Upper tracing - thoracic oesophagus (10.0 cm anterior to LOS); lower tracing - dorsal sac of rumen (10.0 cm posterior to LOS). The lower arrows indicate the onset and termination of reflux. Transient decrease in pressure in the thoracic oesophagus (upper arrows) coincided with deep inspiration were associated with reflux. The ruminal activity was not marked. The pressure changes in the ruminal tracing were associated with the abdominal pressure during breathing.

FIGURE 13.14

Direction of thoracic oesophageal pressure waves during light alphaxalone/alphadolone anaesthesia. Upper tracing - anterior thoracic oesophagus (25.0 cm anterior to LOS); lower tracing - posterior thoracic oesophagus (5.0 cm anterior to LOS). The pressure waves of the anterior thoracic oesophagus preceded those of posterior thoracic oesophagus, i.e. the pressure waves were peristaltic.

FIGURE 13.12

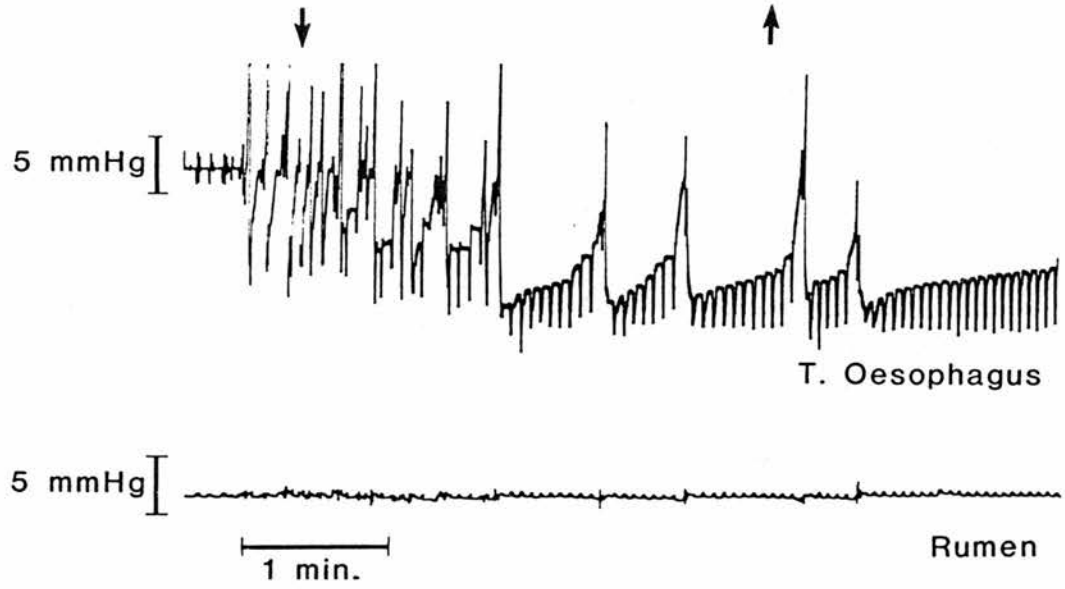


FIGURE 13.13

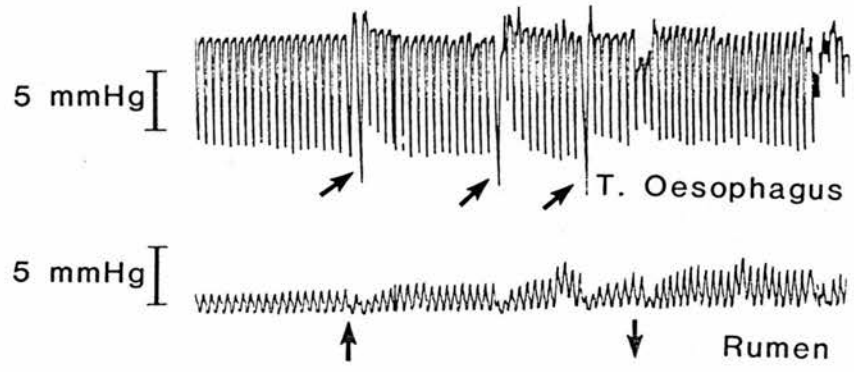


FIGURE 13.14

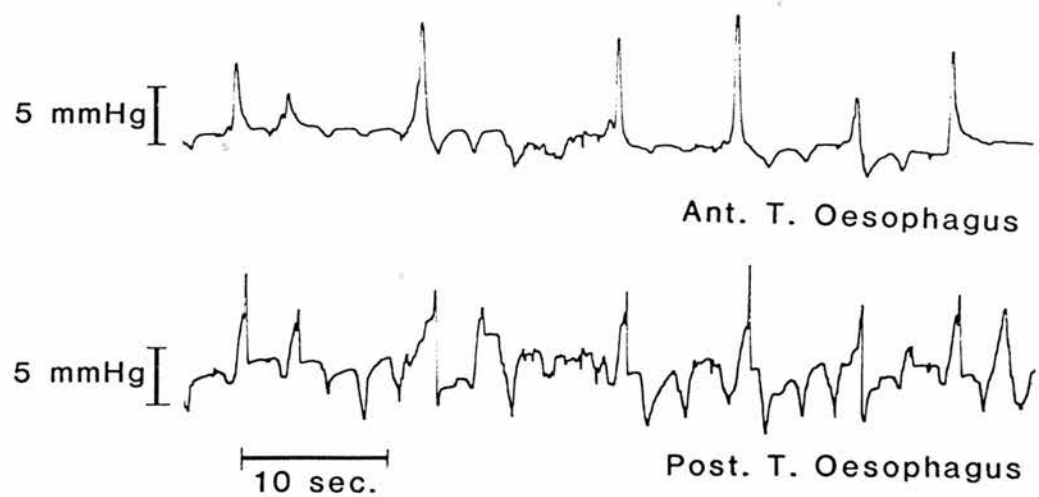
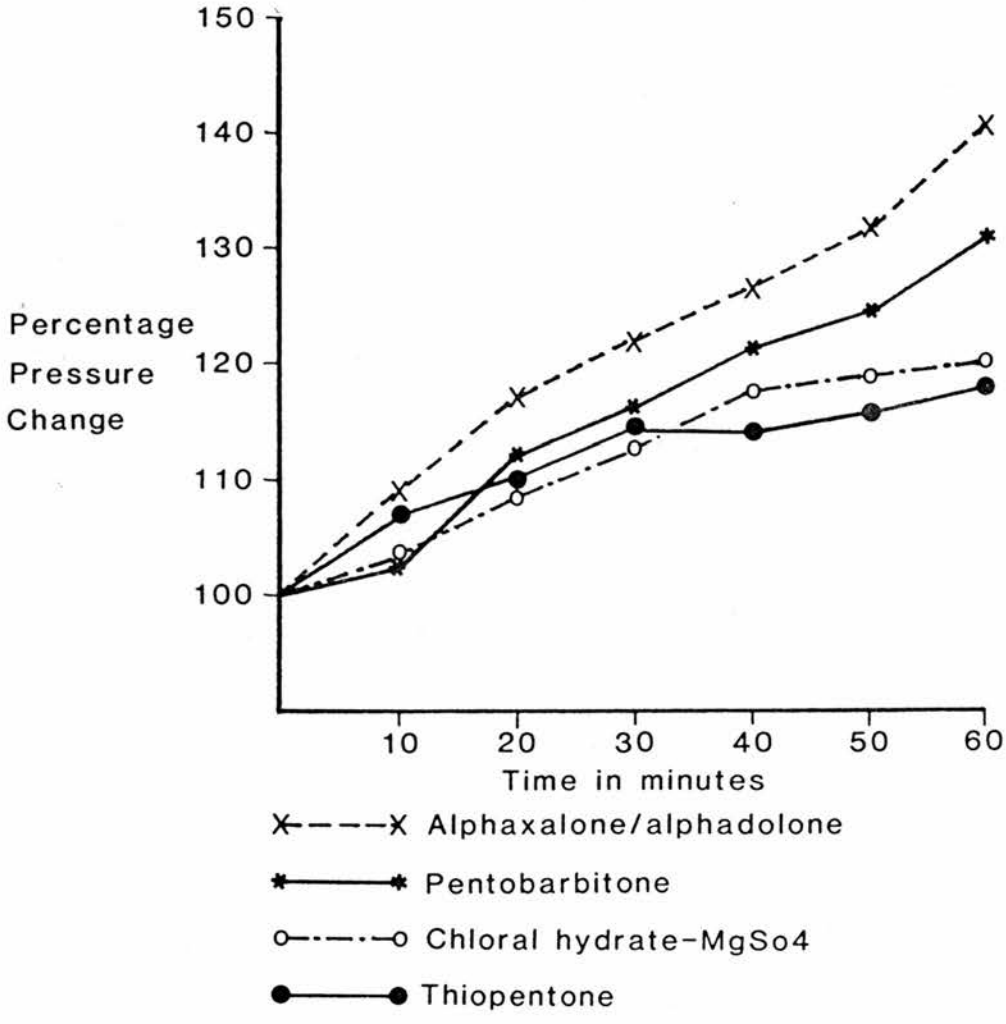


FIGURE 13.15
Relationship of intraruminal pressure built up between alphaxalone/alphadolone, pentobarbitone, thiopentone and chloral hydrate-MgSo4 anaesthesia over a period of 60 minutes.



to the use of starved sheep. Also, the posterior part of the animal was tilted down and on some occasions, the rumen contents were temporarily taken out to empty the rumen. These factors have been found to reduce the incidence of reflux (Chapters 5 and 6). Harrison (1964) did not however, mention the posture of the animal during anaesthesia which also affects refluxing.

Oesophageal and ruminal activities were absent during barbiturate anaesthesia which may be associated with the central inhibition of deglutition and gastric motor centres in the brain stem. Oesophageal manometry did not show any peristaltic waves regardless of the depth of anaesthesia. These waves act as a protective mechanism against reflux. The absence of these waves, therefore, in barbiturate anaesthesia renders the animal more vulnerable to gastro-oesophageal reflux. Harrison (1964) stated that swallowing movements and the reflex control of eructation and regurgitation reappeared as soon as anaesthesia lightened. These observations were not supported here using manometry.

The influence of pentobarbitone on lower oesophageal sphincter pressure has not been investigated in the present studies. The action of pentobarbitone, however, has been studied in anaesthetized opossums by Goyal et al. (1973) who reported that it has no measurable effect on LOS pressure.

Chloral hydrate-$MgSO_4$ and alphaxalone/alphadolone: Chloral hydrate is a traditional anaesthetic agent. It has long been replaced by barbiturates and subsequently by steroid anesthetic agents (e.g. Saffan). Chloral hydrate may produce ventricular fibrillation and sudden death during the recovery period (Hall and Clarke, 1983).

Oesophageal and ruminal activities were present during these anaesthetics when the depth of anaesthesia was relatively light suggesting that these intravenous agents do not suppress the motor centres of these organs. These activities, however, were not so frequent as they were in the case of halothane (Chapter 9). Hall and Clarke (1983) also stated that chloral hydrate has no effect on gastric motility in sheep.

Gastro-oesophageal reflux occurred during anaesthesia produced by all the four intravenous anaesthetic agents. The highest occurrence (70%) was met with thiopentone and the lowest (40%) with alphaxalone/alphadolone anaesthesia. The increased incidence of reflux in the case of thiopentone might be associated with the total abolition of oesophageal peristaltic motility which acts as a defensive mechanism preventing the occurrence of reflux. Alphaxalone/alphadolone appeared to be the best intravenous anaesthetic agent from the standpoint of reflux.

It has been demonstrated in Chapter 9 that increased concentration of halothane progressively diminishes the ruminal pressure waves. In the present studies, chloral hydrate-$MgSO_4$ caused instant abolition of ruminal pressure waves (Figure 13.8). The reason for this may be that, intravenous anaesthetic agents go directly into the circulation and reach the brain more quickly than inhalation anaesthetics.

During pentobarbitone, thiopentone and alphaxalone/alphadolone anaesthesia, the thoracic oesophagus showed a transient reduction in pressure associated with inspiration. This pattern of oesophageal pressure changes might change the pressure gradient across the LOS and may provoke reflux.

The intraruminal pressure built up in alphaxalone/alphadolone anaesthesia was significantly higher than that in thiopentone anaesthesia. The reasons are not clearly understood.

Conclusions

(i) The barbiturates (pentobarbitone, thiopentone) might increase the incidence of gastro-oesophageal reflux, because defensive oesophageal peristalsis is totally abolished.

CHAPTER FOURTEEN

GENERAL DISCUSSION AND CONCLUSIONS

Because of hazards associated with gastro-oesophageal reflux during ruminant anaesthesia, it is accepted by the majority of veterinary anaesthetists that general anaesthesia should be avoided as far as possible where sedation and local analgesia could be used. In a relatively deep plane of anaesthesia, coughing and swallowing reflexes disappear and therefore, when a reflux occurs, the rumen material may be inhaled in the absence of protective mechanisms. Modern anaesthetic machines and agents have made the technique relatively easy and therefore, surgeons and anaesthetists frequently prefer general anaesthesia even for minor surgical interventions. This increasing trend towards the practice of general anaesthesia is also associated with increased risk from the hazards associated with gastro-oesophageal reflux. During the course of the present investigation, five animals died, two deaths occurring on the operating table and three post-operatively in the animal box. At necropsy, the two deaths on the operating table were found to be related to inhalation of rumen material. The other three deaths may have been associated with inhalation of rumen material although this was not determined directly. Therefore, of all the anaesthetics used this constitutes about 1% mortality which may have been related to gastro-oesophageal reflux. No attempt, however, was made to determine the morbidity (% of anaesthetics associated with inhalation of rumen contents) of the hazard, but this may be fairly high. Harrison (1964) reported in sheep a 6% incidence of inhalation associated with pentobarbitone anaesthesia. The total incidence of reflux which occurred during these controlled investigations was 64% and in general those conditions most likely to provoke reflux are:

presence of a second cuffed endotracheal tube in the cervical oesophagus; pre-operative feeding and head down position. Also, in the present investigation the occurrence of reflux was critically monitored and any amount over 5 ml was considered a reflux. This volume of rumen material would probably not be considered a reflux by the anaesthetists.

This investigation demonstrates that:

(1) Management of animals prior to and during anaesthesia influences the incidence of reflux. (a) Starvation of the animal from food and water for 24 hours significantly reduced the incidence of reflux. (b) Reflux was also significantly influenced by the positioning of the animals during anaesthesia. Reflux was more common in head down position than in the head up position.

(2) The use of different anaesthetic agents also influenced the incidence of reflux.

The depth of anaesthesia had no influence on the occurrence of reflux. Manometric studies of the lower oesophageal sphincter (LOS) in the anaesthetized sheep demonstrated that the plane of anaesthesia had no significant influence on its resting pressure (Chapter 10). The light plane of anaesthesia used in the present investigation was probably too light for surgical interference, but was totally adequate for the purposes of restraint. The deep plane of anaesthesia was adequate for surgical purposes. Respiration at this plane of anaesthesia was spontaneous unless it was embarrassed by excessive intraruminal gas production. An incidental finding was that in the case of very deep anaesthesia, as observed during euthanasia, large volumes of rumen material were refluxed just

before death. This may suggest that overdosing of the animal with anaesthetics may have an effect which involves complete relaxation of LOS either directly or due to medullary paralysis.

Anaesthetics greatly influenced the motor control of the oesophagus, LOS and reticulorumen. A sound knowledge of these motor functions during anaesthesia might help understand the mechanism of reflux. Manometric studies in the present investigation demonstrated that in light anaesthesia using halothane, the oesophageal, ruminal and LOS motor functions remain relatively unimpaired. In deep anaesthesia, however, these activities were usually abolished or greatly suppressed except in the case of LOS which still showed rhythmic fluctuation in baseline pressure (Chapter 10). Electromyographic studies of the functions of oesophagus, LOS and reticulorumen also supported the manometric findings.

Intravenous anaesthetic agents also influenced the functional state of these organs. Barbiturates (pentobarbitone and thiopentone) usually caused abolition of oesophageal and ruminal functions in both light and deep planes of anaesthesia. These activities, however, were present in light anaesthesia using chloral hydrate-magnesium sulphate and alaphaxalone/alphadolone.

The mechanism of reflux in the anaesthetized sheep has been studied for the first time in the present investigation. Four hypothetical circumstances were suggested by Leek (1975) whereby rumen material may be refluxed into the oesophagus. These are (1) regurgitation of the cud bolus during rumination, (2) vomiting under certain experimental conditions, (3) eructation of ruminal gases and (4) passive flow of rumen material through the relaxed

oesophageal sphincters. These possibilities can now be discussed in the light of the findings of the present investigation. Firstly, reflux of rumen material during anaesthesia was not associated with regurgitation in these investigations since there was no oesophageal antiperistalsis measured which is invariably present in the case of regurgitation in the conscious sheep. In the present investigation, the oesophageal pressure waves recorded during anaesthesia were always peristaltic in nature even when the animal was producing reflux. Secondly, reflux was not associated with vomiting which usually occurs in monogastric animals whereby the gastric contents are expelled from the mouth associated with retching movements. In ruminants, the true stomach (abomasum) is located far from the oesophagus and during vomiting the abomasal contents would first enter the omasum and then the reticulorumen. Vomiting of abomasal contents into the mouth, therefore, seems very unlikely in the ruminant. The reticuloruminal contents, however, can be vomited under experimental conditions in sheep (Leek, 1975). In the anaesthetized sheep, large volumes of rumen material are sometimes refluxed when they are associated with abdomino-thoracic movement (M.A. Camburn, unpublished observation). Therefore, the abdominal press which is characteristic of vomiting in the monogastric animal is rarely observed in the ruminant. Thirdly, reflux demonstrated here was not associated with classical eructation which again involves rapid oesophageal antiperistalsis. The evidence obtained in the present investigation, however, supports Leek's fourth possibility, i.e. reflux occurs passively in the oesophagus in the absence of oesophageal antiperistalsis. In the case of

passive reflux, there must be some driving mechanism to force the rumen material out. Gastro-oesophageal hydrostatic pressure gradient acts as the driving mechanism for the cranial movement of the rumen material. In conditions where the hydrostatic pressure gradient between the oesophagus and reticulorumen was less, for example in the case of the head up position, the occurrence of reflux was significantly reduced (Chapter 6). The gastro-oesophageal pressure gradient resulting from increased intraruminal gas production also favours the occurrence of reflux. It has been observed (Chapter 7) that the occurrence of reflux is significantly lower in the starved animal than the unstarved. This may be associated with the significantly reduced intraruminal gas production in the starved animal.

The gastro-oesophageal pressure gradient is also a major factor in initiating regurgitation in the conscious ruminant (Dougherty, 1961; Stevens and Sellers, 1968; Phillipson, 1970). The insufflation study in the present investigation, however, showed that this gradient has little effect in producing reflux since the amount of intraruminal pressure required to produce reflux was about five times the pressure that can naturally be built up during anaesthesia lasting for 2-3 hours.

The mechanisms which are involved in the regulation of reflux during anaesthesia are still not clearly understood. The high pressure zone (HPZ) present at the gastro-oesophageal junction in the anaesthetized sheep (Chapter 10), however, appears to play an important role in the regulation of reflux. The pressure at this zone is

usually higher than that in the rumen and thus acts as a barrier to the flow of rumen material up into the oesophagus. In situations where the occurrence of reflux is favoured, for example at the time of reticuloruminal contractions, the barrier pressure is also increased by 4-5 times the amplitude of reticuloruminal contractions. The LOS pressure was also increased when the intraruminal pressure was raised artificially by abdominal compression. These patterns of LOS function are obviously involved in the control mechanism of reflux. The occurrence of reflux, therefore, demands a prior relaxation of the LOS. The manometric studies in the present investigation demonstrated that the occurrence of reflux into the oesophagus is usually associated with the diminution of LOS pressure (Chapter 10). The LOS has also been reported to regulate gastro-oesophageal reflux in man (Fisher, Malmud, Roberts and Lobis, 1977). It would appear that the mechanisms which regulate the LOS might regulate the occurrence of gastro-oesophageal reflux. These are, however, still not clearly understood. In monogastric animals and man, these mechanisms have been reported to involve nervous innervation both extrinsic and intrinsic (Goyal and Cobb, 1981), gastrointestinal hormones (e.g. gastrin, prostaglandins) (Lipshutz and Cohen, 1971), inherent tonicity of the sphincteric smooth muscle (Thomas, 1981). In the present investigation, gastrin analogue pentagastrin, beta blocker (propranolol hydrochloride) and anti-cholinergic drug (atropine sulphate) did not produce any significant influence on LOS pressure suggesting that in ruminants, the LOS control mechanisms might be different from those in monogastric animals. Further investigation is necessary to study the regulatory mechanism of LOS.

The presence of LOS as determined by a high pressure zone in the anaesthetized sheep has been demonstrated in this investigation for the first time using balloon tip catheter and continuous slow pull through methods (Chapter 10). A previous study by Winship et al. (1964) failed to identify the high pressure zone in the conscious sheep using fluid filled open tip catheter and multiple pull through methods. This technique is not suitable to monitor this zone of high pressure as has been observed in the present investigation (Chapter 10). Although the balloon catheters are reliable in identifying the high pressure zone, they produce many inaccuracies particularly in recording the absolute pressure. In the present investigation, the LOS pressures recorded by a balloon tip catheter were significantly greater than those obtained by an open tip catheter. These findings are in agreement with other reports (Pope, 1967; Rinaldo and Levey, 1968; Silber, 1968).

The mechanisms which are involved in reducing the occurrence of reflux in the anaesthetized sheep are the factors which maintain the (1) presence of LOS in the form of a high pressure zone, (2) presence of oesophageal peristaltic waves in light anaesthesia and (3) presence of an upper oesophageal sphincter which acts as a second barrier to the outflow of reflux when the oesophageal peristalsis fails to sweep the materials back to the rumen or when these waves are absent.

From these general considerations, it may be concluded that the occurrence of reflux may be minimised by the following practices:

(1) The animals should be starved of food and water for 24 hours.

(2) It is common practice to keep the head tilted down so that any rumen material from the oesophagus can be drained off along the hydrostatic pressure gradient. Linzell (1964) however, reported that inhalation of rumen contents may occur even if the head is tilted downward. The animal after induction of anaesthesia should be positioned with its head up. This position had the lowest incidence of reflux (Chapter 6).

(3) The collection technique of refluxed rumen material in the present investigation did not allow any rumen material to enter the mouth. This involves the use of a second cuffed endotracheal tube in the cervical oesophagus for collecting the refluxed rumen material. This method can be practised clinically to prevent inhalation of refluxed rumen material into the lungs. The technique, however, does provoke greater incidence of reflux.

(4) A light plane of anaesthesia should be maintained where possible, because during light anaesthesia the protective oesophageal peristalsis mechanism is present and the LOS tone is maintained.

Future Work

The following areas require further investigation:

1. (a) In the present investigation, the influence of total starvation (both food and water) has been studied. Some anaesthetists, however, starve the animals from food only allowing access to water ad libitum. In this condition the rumen contents would be more liquid with increased volume, a condition which may be more vulnerable to the occurrence of reflux during anaesthesia.

(b) Periods of starvation in excess of 24 hours might also influence the occurrence of reflux.

(c) Concentrate feeds produce rapid microbial fermentation (Kölling, 1976) and therefore, it is advisable not to allow any concentrates 48 hours prior to induction of anaesthesia. The advantage of this practice, however, has not been tested in the present investigation.

2. Gastro-oesophageal reflux has been investigated here by manometry, electromyography and direct observation. Radiographic and endoscopic methods may also be employed and these may provide some additional or supporting evidence e.g. length and patency of LOS, the presence of "hidden reflux" in the thoracic oesophagus etc.

3. (a) Because of the importance of LOS in regulating the occurrence of reflux, it is necessary to investigate in detail, the factors which influence the LOS pressure and its control mechanism. These factors, however, are still obscure. Gastrin analogue pentagastrin, atropine and propranolol did not produce any significant influence on LOS pressure. The effect of other gastrointestinal hormones (e.g. prostaglandins) on LOS pressure may be studied.

(b) The influence of extrinsic nerves (vagus and splanchnic) on LOS pressure has not been investigated here. This may be studied more precisely in acute experiments by denervation and electrical excitation. An obvious example is the reflex initiated from the inflated cuff in the cervical oesophagus on the LOS pressure.

(c) In the present investigation, it has been demonstrated that the LOS contraction occurred in association with rumen contraction.

The mechanism of this pattern of LOS contraction is not known but appears to be as a result of vago-vagal reflex. This can also be studied in acute experiments.

4. Electromyographic activity of the gastro-oesophageal junction was studied by implanting electrodes about 1.5 cm posterior to the diaphragm. Because of the difficulties in implanting electrodes at this site and the necessity for the transthoracic approach, these activities may not be related to LOS as the position of the electrodes may not be at the same level of high pressure zone. Further investigation is essential to characterise the LOS function electromyographically and more specifically.

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APPENDICES

Appendix 5.1 Effect of feeding regime on the occurrence of reflux during halothane anaesthesia (unstarved).

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	567E	+	2000
2	KPB1	+	500
3	KPB2	+	5
4	631E	+	1200
5	303E	+	600
6	ACS1	+	5
7	KPB3	+	25
8	567E	+	1715
9	KPB4	+	225
10	KPB5	+	635
11	631E	+	5
12	555E	+	195
13	KPB6	-	-
14	KPB7	+	760
15	567E	+	75
16	510E	+	15
17	555E	+	140
18	KPB8	+	5
19	KPB9	+	715
20	555E	+	90
21	567E	+	80
22	KPB10	+	1420
23	659E	-	-
24	303E	-	-
25	510E	+	10

Appendix 5.2 Effect of feeding regime on the occurrence of reflux during halothane anaesthesia (starved for 24 hours)

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	ACS2	-	-
2	V894	+	40
3	127B	+	105
4	552E	+	3580
5	Z650	+	1725
6	566E	-	-
7	567E	+	1885
8	Z981	+	100
9	566E	-	-
10	Z650	+	530
11	552E	+	40
12	645B	+	5
13	631E	+	200
14	566E	-	-
15	627E	+	90
16	518E	-	-
17	631E	-	-
18	645B	+	335
19	631E	-	-
20	552E	+	25
21	V894	+	360
22	127B	+	225
23	518E	-	-
24	627E	-	-
25	127B	+	15

Appendix 6.1 Dorsal head down position and the occurrence of reflux during halothane anaesthesia

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	517E	+	10
2	518E	+	925
3	Z650	+	65
4	645B	+	410
5	567E	+	350
6	518E	+	700
7	ACS3	+	300
8	ACS4	+	145
9	KPB11	+	260
10	ACS5	+	1950
11	528E	+	865
12	517E	+	350

Appendix 6.2 Right lateral head up position and the occurrence of reflux during halothane anaesthesia

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	V894	-	-
2	X511	-	-
3	552E	-	-
4	X511	+	480
5	303E	-	-
6	V894	-	-
7	303E	-	-
8	524E	-	-
9	303E	-	-
10	X511	-	-
11	521E	-	-
12	X511	+	245
13	622E	-	-
14	524E	-	-
15	521E	-	-
16	552E	-	-
17	V894	-	-
18	524E	-	-
19	518E	-	-
20	524E	-	-
21	552E	-	-
22	V894	-	-
23	518E	-	-
24	303E	+	125

Appendix 6.3 Right lateral head down position and the occurrence of reflux during halothane anaesthesia

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	567E	+	2000
2	KPB1	+	500
3	KPB2	+	5
4	631E	+	1200
5	303E	+	600
6	ACS1	+	5
7	KPB3	+	25
8	567E	+	1715
9	KPB4	+	225
10	KPB5	+	635
11	631E	+	5
12	555E	+	195
13	KPB6	-	-
14	KPB7	+	760
15	567E	+	75
16	510E	+	15
17	555E	+	140
18	KPB8	+	5
19	KPB9	+	715
20	555E	+	90
21	567E	+	80
22	KPB10	+	1420
23	659E	-	-
24	303E	-	-
25	510E	+	10

Appendix 6.4 Left lateral head down position and the occurrence of reflux during halothane anaesthesia

Anaesthetic number		Occurrence of reflux	Quantity of reflux (ml)
1	567E	-	-
2	ACS6	+	155
3	KPB12	+	315
4	KPB13	+	80
5	622E	+	25
6	ACS7	+	485
7	X511	+	160
8	V894	-	-
9	631E	-	-
10	645B	+	25
11	517E	-	-
12	V864	-	-
13	182E	+	25
14	631E	-	-

Appendix 6.5 Right lateral head down position (without cuffed endotracheal tube in the cervical oseophagus) and the occurrence of reflux during halothane anaesthesia

Anaesthetic number		Occurrence of reflux	Quantity of reflux and saliva (ml)
1	515E	-	100
2	631E	+	880
3	182E	+	1230
4	V864	+	220
5	127B	+	1530
6	515E	-	105
7	631E	-	275
8	182E	-	25
9	V864	-	215
10	515E	+	50
11	515E	+	85

Appendix 7.1 Individual values of intraruminal pressure built up during halothane anaesthesia (light followed by deep)

Sheep No.	Initial pressure (mmHg)	Pressure after 10 mins (mmHg)	Pressure after 20 mins (mmHg)	Pressure after 30 mins (mmHg)	Pressure after 40 mins (mmHg)	Pressure after 50 mins (mmHg)	Pressure after 60 mins (mmHg)
	8.0	9.0	9.5	10.5	12.5	11.0	14.0
192D	5.0	5.5	5.0	6.0	6.0	7.0	7.0
	5.5	6.5	7.0	7.5	7.0	7.0	7.5
Mean	6.16	7.0	7.16	8.0	8.5	8.33	9.5
	5.5	6.5	8.0	8.0	7.0	8.0	9.5
243D	4.0	3.5	4.0	4.5	5.0	7.0	8.5
	7.5	8.0	8.0	8.0	8.5	10.5	10.5
Mean	5.66	6.0	6.66	6.83	6.83	8.5	9.5
	13.0	14.0	14.0	15.0	15.0	14.0	12.5
27x91	10.0	13.1	13.0	14.0	14.8	15.4	16.5
	3.0	3.0	3.5	3.5	4.5	4.0	4.0
Mean	8.66	10.03	10.16	10.83	11.43	11.13	11.0
	6.0	6.0	8.0	8.5	6.5	9.0	8.0
289D	11.0	11.0	11.0	11.0	11.0	11.0	11.0
	3.0	3.0	3.0	3.0	3.5	3.0	3.0
Mean	6.66	6.66	7.33	7.5	7.0	7.66	7.33
Total Mean	6.78	7.42	7.82	8.29	8.44	8.90	9.33
S.D.	1.31	1.78	1.58	1.75	2.12	1.52	1.51

Appendix 7.2 Individual values of intraruminal pressure built up during halothane anaesthesia (deep followed by light)

Sheep No.	Initial pressure (mmHg)	Pressure after 10 mins (mmHg)	Pressure after 20 mins (mmHg)	Pressure after 30 mins (mmHg)	Pressure after 40 mins (mmHg)	Pressure after 50 mins (mmHg)	Pressure after 60 mins (mmHg)
	6.0	9.0	10.5	13.0	17.0	16.0	14.0
192D	6.0	9.5	10.8	15.0	12.0	12.0	12.0
	7.5	8.0	8.5	9.0	10.5	12.5	12.5
Mean	6.5	8.83	9.93	12.33	13.16	13.5	12.83
	7.0	8.5	11.2	11.5	11.5	12.5	12.5
243D	2.5	2.5	5.5	7.0	8.5	10.0	12.5
	7.5	7.5	8.0	9.0	9.0	9.0	10.0
Mean	5.66	6.16	8.23	9.16	9.66	10.5	11.66
	13.0	14.0	14.0	17.0	19.0	21.0	24.0
27x91	5.0	5.0	5.5	6.0	6.0	6.0	8.0
	10.0	10.5	10.5	10.0	12.0	12.5	12.5
Mean	9.33	9.83	10.0	11.0	12.33	13.16	14.83
	6.0	6.0	7.5	8.5	7.5	8.5	7.0
289D	7.0	8.5	11.0	11.5	13.0	15.0	15.0
	8.0	9.0	9.5	10.0	10.0	11.0	11.5
Mean	7.0	7.83	9.33	10.0	10.16	11.5	11.16
Total Mean	7.62	8.16	9.37	10.61	11.32	12.16	12.62
S.D.	1.57	1.56	0.81	1.36	1.68	1.41	1.63

Appendix 7.3 Maximum rumeno-oesophageal pressure gradient and the occurrence of reflux during all planes of halothane anaesthesia

Sheep No.	Maximum Pressure Gradient (mmHg)			Occurrence of reflux
	Rumen	Thoracic oseophagus	Gradient	
192D				
1	12.0	-1.5	13.5	+
2	15.5	3.5	12.0	+
3	7.0	4.5	2.5	+
4	12.5	1.5	11.0	+
5	12.5	3.5	13.0	-
6	7.0	0.0	7.0	-
243D				
1	8.0	3.0	5.0	-
2	12.0	1.0	11.0	+
3	8.5	1.0	7.5	-
4	8.5	2.5	6.0	-
5	8.0	2.5	5.5	-
6	10.0	0.0	10.0	-
27x91				
1	8.5	1.0	7.5	-
2	11.5	5.5	6.0	-
3	4.5	2.0	2.5	+
4	10.0	1.5	8.5	-
5	9.0	0.0	9.0	-
6	10.0	0.0	10.0	-
289D				
1	9.0	1.0	8.0	-
2	8.0	2.5	5.5	-
3	11.0	1.0	10.0	+
4	15.0	3.5	11.5	+
5	8.0	-2.0	10.0	+
6	3.0	0.5	2.5	-

Appendix 8.1 Intraruminal insufflation and the pressure threshold producing LOS patency during light and deep planes of anaesthesia

Sheep Number	Intraruminal Threshold Pressure Producing LOS Patency (mm Hg)											
	Light Plane					Mean	Deep Plane					Mean
Black tag	24.0	54.0	56.0	54.0	60.0	49.6	60.0	54.0	52.0	55.0	50.0	54.2
	39.0	49.0	45.0	30.0	30.0	38.6	20.0	30.0	42.0	16.0	18.0	25.2
	48.0	40.0	30.0	36.0	37.0	38.2	28.0	26.0	26.0	35.0	30.0	29.0
230D	44.0	50.0	43.0	60.0	65.0	52.4	47.0	45.0	55.0	50.0	48.0	49.0
	40.0	50.0	40.0	44.0	40.0	42.8	20.0	20.0	26.0	26.0	25.0	23.4
	30.0	25.0	37.0	28.0	41.0	32.2	20.0	25.0	19.0	25.0	30.0	23.8
243D	37.0	36.0	45.0	32.0	36.0	37.2	50.0	32.0	42.0	22.0	25.0	34.2
	40.0	45.0	50.0	40.0	35.0	42.0	30.0	40.0	30.0	35.0	40.0	35.0
	30.0	40.0	35.0	40.0	20.0	33.0	30.0	48.0	24.0	45.0	24.0	34.2
27x91	31.0	30.0	55.0	31.0	42.0	37.8	55.0	32.0	42.0	45.0	34.0	41.6
	40.0	60.0	40.0	25.0	45.0	42.0	35.0	22.0	28.0	32.0	34.0	30.2
	35.0	30.0	30.0	10.0	45.0	30.0	22.0	26.0	30.0	25.0	20.0	24.6
Total mean						39.65						33.7
S.D.						6.66						10.05

Appendix 9.1 Duration, amplitude and velocity of thoracic oesophageal pressure waves associated with reflux in light anaesthesia

Sheep 271D

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.8	7.5	1.0	7.5	33.33
2	0.8	15.0	1.2	9.0	16.66
3	0.7	10.0	1.2	7.5	25.0
4	0.8	7.5	1.2	10.0	25.0
5	0.5	7.5	1.3	10.0	33.33
6	0.9	7.5	1.2	10.0	33.33
7	0.5	11.5	1.0	9.5	25.0
8	0.6	7.5	1.0	10.0	25.0
9	0.8	11.0	1.2	9.0	25.0
10	0.5	7.5	1.2	8.0	20.0
11	0.7	10.5	1.2	12.0	20.0
12	0.8	12.5	1.0	8.0	16.66
13	0.6	5.5	1.2	7.5	25.0
14	0.8	13.0	1.0	7.5	25.0
15	1.0	5.5	1.2	7.5	33.33
16	1.0	11.0	1.0	7.5	25.0
17	0.7	10.0	1.0	8.0	33.33
18	1.0	9.5	1.2	9.0	25.0
19	1.0	10.0	1.5	9.0	25.0
20	0.9	10.5	1.2	8.0	33.33
21	1.0	11.0	1.0	10.0	20.0
22	1.0	10.0	2.0	7.0	25.0
23	1.0	10.5	1.2	5.5	33.33
24	0.9	11.0	1.2	8.0	20.0
25	0.8	9.0	1.2	8.5	25.0
Mean	0.8	9.68	1.18	8.54	25.86
S.D.	0.16	2.28	0.2	1.37	5.42

Appendix 9.1 (continued)

Sheep 127B

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.8	9.5	1.4	10.0	33.33
2	0.9	10.5	1.4	6.0	33.33
3	0.6	5.0	1.6	8.0	25.0
4	0.8	15.0	1.6	6.0	25.0
5	0.8	15.0	1.7	6.0	25.0
6	0.6	15.0	0.9	7.5	20.0
7	1.2	11.0	1.3	7.5	33.33
8	1.2	7.5	1.6	7.5	25.0
9	2.0	16.0	2.0	6.0	20.0
10	1.6	12.5	2.0	5.0	20.0
11	1.5	10.0	1.2	11.5	20.0
12	0.8	18.5	1.2	9.0	25.0
13	0.8	11.0	0.8	8.5	25.0
14	1.2	8.0	0.8	10.0	25.0
15	1.2	12.5	0.8	8.0	20.0
16	0.5	3.5	2.8	11.0	20.0
17	1.2	10.0	2.4	5.0	33.33
18	0.7	8.5	2.0	5.0	20.0
19	3.2	13.0	2.0	7.5	33.33
20	1.6	12.5	1.6	8.5	33.33
21	0.6	12.0	0.8	7.5	25.0
22	4.0	12.5	0.8	7.5	25.0
23	3.2	15.0	1.6	6.0	25.0
24	1.4	10.0	0.9	7.5	25.0
25	1.2	20.0	1.2	7.5	25.0
Mean	1.34	11.76	1.45	7.58	25.59
S.D.	0.89	3.82	0.54	1.77	4.92

Appendix 9.1 (continued)

Sheep 518E

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.8	12.5	1.6	10.5	33.33
2	1.5	5.0	1.2	7.5	25.0
3	1.2	6.5	2.0	15.0	25.0
4	1.6	17.0	2.8	10.0	33.33
5	1.2	17.0	1.3	6.0	33.33
6	0.4	14.0	1.6	12.5	33.33
7	1.2	7.5	2.2	11.0	25.0
8	1.0	15.0	1.6	7.0	25.0
9	0.8	10.0	1.2	8.0	25.0
10	1.2	8.5	2.0	6.5	33.33
11	0.8	11.0	2.0	10.0	33.33
12	1.2	9.5	1.3	7.5	33.33
13	3.2	13.5	4.4	7.5	33.33
14	1.2	10.0	1.6	5.0	25.0
15	1.6	24.0	1.6	10.0	25.0
16	1.6	25.0	1.6	11.0	25.0
17	2.2	25.0	2.2	11.0	33.33
18	0.8	5.0	1.6	10.0	16.66
19	1.3	15.0	1.2	8.0	20.0
20	0.8	7.0	0.8	7.0	25.0
21	3.2	17.0	4.0	11.0	20.0
22	1.6	5.0	1.6	7.5	25.0
23	0.6	5.0	1.5	4.0	25.0
24	1.2	15.0	2.8	10.0	25.0
25	1.0	15.0	2.0	7.0	25.0
Mean	1.32	12.6	1.9	8.82	27.26
S.D.	0.68	6.08	0.83	2.49	5.06

Appendix 9.1 (continued)

Sheep Z981

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	1.2	12.0	1.6	6.5	25.0
2	0.8	7.5	2.0	7.0	20.0
3	1.6	12.5	1.6	7.5	25.0
4	1.6	11.0	1.8	7.0	25.0
5	1.2	12.0	2.0	6.5	20.0
6	0.8	12.0	1.6	7.5	25.0
7	0.8	11.0	1.2	7.5	33.33
8	1.2	12.5	2.0	7.5	20.0
9	0.8	12.5	1.6	7.5	20.0
10	1.2	12.5	1.6	7.0	25.0
11	0.8	10.0	1.6	7.5	25.0
12	1.0	10.0	1.5	6.5	25.0
13	0.8	9.0	1.6	7.5	25.0
14	1.2	9.5	1.6	12.5	25.0
15	1.2	9.0	1.6	12.0	25.0
16	1.2	8.0	1.0	10.0	25.0
17	0.8	10.0	1.2	8.0	25.0
18	0.8	12.0	1.6	6.0	25.0
19	1.2	10.0	1.2	8.0	25.0
20	0.8	9.0	2.0	8.0	25.0
21	1.0	10.0	2.0	8.0	25.0
22	0.8	8.0	1.6	9.0	20.0
23	0.8	8.0	1.6	8.0	25.0
24	0.8	9.0	1.8	7.0	20.0
25	1.2	10.0	1.6	9.5	20.0
Mean	1.02	10.28	1.62	7.96	23.93
S.D.	0.25	1.62	0.26	1.58	2.99

Appendix 9.1 (continued)

Sheep 645B

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.8	13.0	0.8	10.0	50.0
2	0.8	12.0	1.0	12.0	33.33
3	0.8	9.0	0.6	7.0	33.33
4	1.2	13.0	1.6	7.5	25.0
5	1.2	15.0	2.0	12.0	50.0
6	0.8	7.5	0.8	7.5	25.0
7	0.8	15.0	2.4	12.0	25.0
8	1.2	17.0	1.6	11.0	50.0
9	1.0	20.0	2.0	15.0	25.0
10	0.8	5.0	1.6	17.0	25.0
11	1.0	15.0	2.0	15.0	25.0
12	0.4	11.0	0.5	7.5	16.66
13	0.5	9.5	1.6	13.0	25.0
14	1.2	13.0	0.5	14.5	25.0
15	1.2	7.5	1.2	11.0	25.0
16	0.4	5.0	2.0	11.0	20.0
17	0.8	19.0	1.2	17.0	25.0
18	0.4	5.0	1.6	13.0	25.0
19	0.8	13.0	1.6	13.5	20.0
20	1.0	12.0	1.2	14.0	20.0
21	0.5	10.0	1.2	14.5	25.0
22	0.8	5.0	1.2	15.0	25.0
23	0.8	3.0	2.0	17.5	20.0
24	0.8	3.0	2.0	22.0	20.0
25	0.8	3.0	1.2	20.0	25.0
Mean	0.83	10.42	1.41	13.18	27.33
S.D.	0.25	5.02	0.53	3.81	9.29

Appendix 9.1 (continued)

Sheep 566E

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	1.4	10.0	1.2	7.5	50.0
2	1.2	14.0	1.6	8.0	33.33
3	1.2	15.0	1.2	8.0	50.0
4	1.0	12.5	1.2	7.5	50.0
5	0.8	14.0	1.2	7.5	50.0
6	1.6	15.0	1.2	7.5	50.0
7	1.0	10.0	1.0	7.5	50.0
8	1.4	15.0	1.6	8.0	25.0
9	1.2	12.5	1.2	7.5	33.33
10	1.4	11.0	1.2	7.0	33.33
11	1.2	13.5	1.0	7.5	25.0
12	2.4	20.0	1.2	7.5	33.33
13	1.6	20.0	1.0	7.5	25.0
14	2.4	17.0	2.0	8.5	33.33
15	1.8	15.0	1.2	8.0	25.0
15	1.6	15.0	2.0	8.5	25.0
17	1.2	15.0	1.0	7.0	33.33
18	1.6	15.0	1.2	7.0	25.0
19	1.2	15.0	1.5	7.5	33.33
20	1.4	15.0	1.2	7.0	50.0
21	1.4	15.0	1.0	7.0	33.33
22	1.4	12.5	1.6	7.5	50.0
23	1.6	17.5	1.2	8.0	25.0
24	1.2	15.0	1.6	7.5	50.0
25	1.2	14.0	1.2	7.5	33.33
Mean	1.41	14.54	1.3	7.58	36.99
S.D.	0.37	2.45	0.28	0.42	10.5

Appendix 9.2 Duration, amplitude and velocity of thoracic oesophageal pressure waves not associated with reflux in light anaesthesia

Sheep Z981

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	1.2	15.0	2.0	15.0	25.0
2	1.2	16.0	2.4	17.5	50.0
3	1.2	16.0	2.0	15.5	25.0
4	1.6	17.0	1.6	11.0	20.0
5	0.8	5.0	2.4	15.0	20.0
6	1.2	16.0	3.2	17.5	50.0
7	0.8	7.0	1.0	18.0	20.0
8	1.2	18.0	1.0	16.0	20.0
9	1.2	8.0	0.8	18.0	20.0
10	1.2	20.0	0.9	17.0	20.0
11	1.4	23.0	0.9	15.0	25.0
12	0.6	18.0	1.3	10.0	20.0
13	0.6	9.0	1.2	14.0	20.0
14	1.4	33.0	1.2	17.0	25.0
15	0.8	18.0	1.2	19.0	25.0
16	0.6	13.0	2.5	16.0	50.0
17	0.8	5.0	2.0	20.0	20.0
18	0.8	29.0	2.4	19.0	20.0
19	1.0	8.0	1.2	18.0	25.0
20	1.4	16.0	1.2	15.0	20.0
21	1.6	6.0	1.2	12.0	16.66
22	1.4	3.0	1.2	12.0	16.66
23	0.8	6.0	1.2	17.0	33.33
24	0.8	21.0	1.2	19.0	25.0
25	1.4	38.0	4.0	20.0	25.0
Mean	1.08	15.36	1.64	16.14	25.46
S.D.	0.31	8.92	0.79	2.73	9.90

Appendix 9.2 (continued)

Sheep 127B

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.9	7.5	1.2	10.0	25.0
2	1.6	9.0	1.2	10.0	33.33
3	1.2	5.5	1.3	10.0	25.0
4	0.8	15.5	1.2	9.0	50.0
5	0.4	8.0	1.2	10.0	25.0
6	0.9	10.0	1.3	10.0	25.0
7	0.5	12.5	1.6	10.0	25.0
8	0.8	15.0	1.2	11.5	20.0
9	2.0	10.0	1.2	11.0	20.0
10	0.6	11.0	1.2	12.5	20.0
11	1.2	5.5	0.8	13.5	25.0
12	0.4	11.0	1.2	10.5	25.0
13	2.0	19.5	1.6	10.0	20.0
14	1.3	22.0	2.0	10.5	20.0
15	0.8	6.0	1.4	15.0	25.0
16	0.5	7.5	0.8	11.0	25.0
17	0.6	7.5	1.6	10.5	20.0
18	0.6	12.0	2.0	8.5	25.0
19	0.8	11.0	1.5	8.0	20.0
20	0.8	2.5	1.2	7.5	20.0
21	0.4	6.0	1.2	5.0	25.0
22	0.5	7.5	2.4	14.0	50.0
23	0.5	5.0	1.3	8.0	25.0
24	0.8	11.0	1.5	17.0	16.0
25	0.8	12.5	1.2	10.0	20.0
Mean	0.86	10.02	1.37	10.52	25.17
S.D.	0.45	4.53	0.35	2.49	8.22

Appendix 9.2 (continued)

Sheep 518E

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.8	15.0	1.4	16.0	25.0
2	0.8	13.0	1.6	17.0	25.0
3	1.2	7.5	2.0	10.5	25.0
4	1.2	7.5	1.6	10.0	25.0
5	0.8	10.0	1.4	10.5	25.0
6	1.2	4.0	2.4	16.5	33.33
7	0.8	2.5	2.0	16.0	50.0
8	0.8	3.5	2.0	17.0	25.0
9	1.4	17.0	2.4	18.0	25.0
10	0.8	18.5	2.2	17.5	20.0
11	0.8	18.5	1.3	20.0	20.0
12	0.8	11.0	1.6	12.5	25.0
13	0.6	7.5	1.3	13.0	25.0
14	0.8	12.0	1.6	16.0	25.0
15	0.5	4.0	2.0	17.0	25.0
16	0.8	20.0	1.6	17.5	25.0
17	0.8	5.5	1.7	14.0	25.0
18	0.8	5.0	1.8	11.0	50.0
19	0.8	6.0	2.0	17.0	33.33
20	0.4	4.0	2.0	16.5	33.33
21	0.8	7.5	2.4	20.0	33.33
22	0.5	21.0	0.8	9.0	25.0
23	0.8	7.5	2.4	19.0	25.0
24	1.2	14.0	2.0	15.0	33.33
25	0.5	5.0	2.0	15.5	33.33
Mean	0.82	9.88	1.82	15.28	28.59
S.D.	0.24	5.74	0.40	3.14	7.61

Appendix 9.2 (continued)

Sheep 566E

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	1.6	4.0	1.2	14.5	25.0
2	1.6	7.0	2.0	17.5	16.66
3	0.4	4.5	2.0	18.0	20.0
4	2.0	7.5	0.4	2.5	25.0
5	0.8	5.0	2.0	17.5	12.5
6	1.0	5.0	1.2	5.0	16.66
7	1.2	4.5	1.0	17.5	25.0
8	2.0	2.5	0.8	18.0	16.66
9	1.2	6.5	2.4	13.5	25.0
10	2.0	7.5	2.0	17.5	16.66
11	1.6	7.0	2.4	17.5	40.0
12	0.8	7.0	0.8	18.0	33.33
13	1.5	15.5	2.0	17.5	20.0
14	0.8	3.5	2.0	18.0	16.66
15	0.6	4.0	1.2	15.0	25.0
16	1.2	6.0	1.0	16.5	50.0
17	1.2	17.5	1.0	18.0	25.0
18	0.8	3.0	0.8	19.0	25.0
19	1.8	3.5	0.8	20.0	50.0
20	0.6	5.0	2.4	19.0	20.0
21	0.4	8.0	0.6	14.0	33.33
22	0.8	3.5	1.0	22.0	33.33
23	0.8	5.0	2.0	40.0	16.66
24	0.8	5.5	0.8	20.0	33.33
25	0.8	5.0	0.8	20.0	25.0
Mean	1.13	6.12	1.38	17.44	25.83
S.D.					

Appendix 9.2 (continued)

Sheep 645B

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	0.6	0.5	1.2	14.5	25.0
2	0.5	1.5	0.8	14.5	25.0
3	0.4	3.0	1.6	14.5	25.0
4	0.6	4.0	1.8	14.0	20.0
5	0.6	4.5	1.0	14.0	25.0
6	0.4	2.5	1.6	14.0	25.0
7	0.8	4.0	2.0	14.5	25.0
8	1.0	3.0	2.0	14.5	33.33
9	0.6	3.5	2.0	15.0	50.0
10	0.8	4.0	2.4	19.0	25.0
11	0.8	3.5	1.2	19.0	33.33
12	0.8	21.0	2.8	15.0	20.0
13	1.0	18.5	1.6	17.0	25.0
14	0.8	23.0	1.0	17.0	40.0
15	0.8	5.5	0.8	14.0	50.0
16	0.8	3.5	1.0	14.5	25.0
17	0.4	10.5	1.8	13.5	20.0
18	0.4	22.0	2.0	13.0	25.0
19	0.6	3.0	1.2	12.5	25.0
20	0.5	23.0	1.0	8.0	33.33
21	0.8	22.5	1.2	12.5	12.0
22	0.6	7.5	1.6	12.5	25.0
23	0.8	22.5	1.2	12.0	33.33
24	0.8	22.5	1.2	13.0	33.33
25	0.8	23.0	1.6	17.0	25.0
Mean	0.68	10.66	1.50	14.36	28.14
S.D.	0.18	8.88	0.51	2.29	8.70

Appendix 9.2 (continued)

Sheep Z650

Pressure waves	Anterior thoracic oesophagus		Posterior thoracic oesophagus		Wave velocity (cm/sec)
	Duration (sec)	Amplitude (mm Hg)	Duration (sec)	Amplitude (mm Hg)	
1	1.0	20.0	1.6	17.5	25.0
2	0.5	10.0	1.2	16.0	25.0
3	0.3	16.0	1.2	15.0	25.0
4	1.0	17.5	1.2	25.0	25.0
5	0.4	25.0	1.2	16.5	20.0
6	1.2	25.0	2.0	19.0	20.0
7	0.4	20.0	1.0	17.5	20.0
8	0.8	17.5	0.8	13.0	16.66
9	0.4	17.5	1.0	13.0	25.0
10	0.8	17.5	0.8	20.0	25.0
11	0.4	19.0	1.0	16.0	20.0
12	0.4	20.0	0.4	10.0	20.0
13	0.8	20.0	0.8	12.5	25.0
14	0.8	25.0	0.8	11.5	25.0
15	0.8	22.0	0.8	11.5	25.0
16	0.4	20.0	1.2	20.0	20.0
17	1.0	25.0	2.0	20.0	33.33
18	0.8	5.0	1.2	15.5	50.0
19	0.4	10.0	1.2	20.0	33.33
20	1.2	22.0	1.2	12.5	25.0
21	0.8	5.0	1.2	16.0	25.0
22	0.8	20.0	1.0	20.0	20.0
23	0.5	15.0	0.8	20.0	20.0
24	0.4	20.0	0.8	15.5	25.0
25	0.8	22.0	0.8	13.0	33.33
Mean	0.68	18.24	1.08	16.26	25.06
S.D.	0.27	5.59	0.36	3.64	6.77

Appendix 9.3 Thoracic oesophageal responses to balloon distension (with 25 cc air) in the cervical oesophagus (10.0 cm anterior to thoracic inlet) during light and deep planes of anaesthesia

Sheep No.	Tone of anterior thoracic oesophagus		Tone of posterior thoracic oesophagus	
	Light plane	Deep plane	Light plane	Deep plane
566E	+	+	+	+
	+	+	+	0
	+	+	+	+
	+	+	+	+
	+	+	+	+
127B	+	+	+	+
	+	+	+	+
	+		+	
	+		+	
	+		+	
645B	-	0	0	+
	-	0	0	0
	0		0	
	0		0	
	0		0	
	0		0	
271E	+	0	+	+
	+	0	+	0
	+	0	+	0
	+	0	+	0
	+	0	0	0
+Increase in tone (%)	71.42 (100)	50 (41.75)	66.66 (93.33)	57.14 (58.35)
-Decrease in tone (%)	9.52 (0.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)
0 No change in tone (%)	19.04 (0.0)	50.0 (58.35)	33.33 (6.67)	42.9 (41.75)

Note: The values in brackets represent the percentages without taking into account sheep 645B.

Appendix 9.4 Cervical and anterior thoracic oesophageal responses to balloon distension (with 25 cc air) in the caudal oesophagus (within 10 cm of LOS) during light and deep planes of anaesthesia

Sheep No.	Tone of cervical oesophagus		Tone of anterior thoracic oesophagus	
	Light plane	Deep plane	Light plane	Deep plane
566E	0	0	+	+
	+	+	+	+
	+	+	+	+
	+	+	+	+
	+	+	+	+
518E	-	-	+	+
	-	-	+	-
	0	-	+	-
	0	-	+	0
	0		-	
	0		-	
	0		-	
	0		0	
645B	0	0	0	+
	0	0	0	+
	0	0	0	+
	0	0	0	+
	0	0	0	+
+Increase in tone (%)	22.2 (30.77)	28.6 (44.4)	50.0 (69.23)	78.6 (66.67)
-Decrease in tone (%)	11.1 (15.38)	28.6 (44.4)	16.7 (23.08)	14.3 (22.22)
0 No change in tone (%)	66.7 (53.85)	42.9 (11.2)	42.9 (7.69)	7.1 (11.11)

Appendix 11.1 Effect of pentagastrin (6.0 $\mu g.kg^{-1}$, i.v.) on lower oesophageal sphincter pressure in anaesthetized sheep

Sheep No.	Injection	Basal LOS pressure (mmHg) n = 3	LOS Pressures After Pentagastrin Injection (mmHg)						
			1 min n = 3	5 mins n = 3	10 mins n = 3	15 mins n = 3	20 mins n = 3	25 mins n = 3	30 mins n = 3
621E	Saline	24.33	24.5	25.33	26.5	27.5	28.0	28.33	28.0
	Pentagastrin	27.0	29.5	29.0	32.0	32.33	35.0	35.66	35.66
517E	Saline	22.16	22.16	22.50	21.50	21.16	21.0	21.66	22.0
	Pentagastrin	23.66	23.66	23.50	23.83	24.83	25.66	26.0	25.50
594E	Saline	22.83	22.66	23.66	24.83	26.66	26.33	25.66	27.33
	Pentagastrin	25.0	27.33	27.0	27.0	25.33	23.66	24.33	24.33
Mean	Saline	23.1	23.1	23.83	24.27	25.1	25.11	25.21	25.77
S.D.		1.11	1.23	1.42	2.54	3.44	3.65	3.35	3.28
Mean	Pentagastrin	25.22	26.83	26.5	27.61	27.49	28.1	28.66	28.49
S.D.		1.68	2.95	2.78	4.11	4.19	6.05	6.11	6.23

Appendix 11.2 Effect of pentagastrin (12.0 $\mu g.kg^{-1}$, i.v.) on lower oesophageal sphincter pressure in anaesthetized sheep

Sheep No.	Injection	Basal LOS pressure (mmHg)	LOS Pressures After Pentagastrin Injection (mmHg)						
			1 min	5 mins	10 mins	15 mins	20 mins	25 mins	30 mins
604E	Saline	14.0	15.5	15.0	14.0	13.5	16.0	15.0	13.0
	Pentagastrin	13.0	15.0	16.0	16.0	16.0	16.5	15.0	12.0
498E	Saline	25.0	26.0	26.0	25.5	25.0	26.0	27.0	28.5
	Pentagastrin	28.0	30.0	30.0	30.0	31.0	29.5	32.5	32.0
506E	Saline	13.0	13.0	11.5	10.5	13.5	25.0	18.0	15.0
	Pentagastrin	15.0	16.0	21.0	17.5	16.0	21.0	25.0	18.5
510E	Saline	25.5	25.5	25.0	24.0	21.0	21.5	22.5	24.5
	Pentagastrin	24.5	24.5	22.0	24.0	25.0	25.0	25.0	25.0
528E	Saline	15.0	14.5	16.0	15.0	17.0	16.0	16.5	15.0
	Pentagastrin	15.0	15.0	15.5	17.0	17.5	17.0	19.0	18.0
Mean	Saline	18.5	18.9	18.7	17.8	18.0	20.9	19.8	19.1
S.D.		6.20	6.31	6.43	6.58	4.98	4.77	4.9	6.76
Mean	Pentagastrin	19.1	20.1	20.9	20.9	21.1	21.8	23.3	21.1
S.D.		6.69	6.82	5.85	5.98	6.67	5.5	6.66	7.63

Appendix 11.3 Effect of pentagastrin (24.0 $\mu g.kg^{-1}$, i.v.) on lower oesophageal sphincter pressure in anaesthetized sheep

Sheep No.	Injection	Basal LOS pressure (mmHg)	LOS Pressures After Pentagastrin Injection (mmHg)						
			1 min	5 mins	10 mins	15 mins	20 mins	25 mins	30 mins
510E	Saline	18.0	18.0	20.0	20.0	21.0	21.0	21.0	21.5
	Pentagastrin	21.0	22.0	23.0	22.5	23.0	24.0	22.0	22.5
502E	Saline	20.0	20.0	22.0	21.5	24.5	27.0	27.5	22.0
	Pentagastrin	27.0	30.0	30.0	32.0	32.5	33.0	34.5	34.0
505E	Saline	28.0	27.5	27.0	29.0	29.0	28.0	30.0	30.0
	Pentagastrin	32.0	32.0	33.5	34.5	35.0	35.0	35.5	36.0
604E	Saline	9.0	9.5	9.0	9.0	9.5	9.5	10.0	10.0
	Pentagastrin	10.5	15.0	13.5	14.0	16.5	17.0	18.5	16.0
506E	Saline	11.0	11.0	8.0	15.0	16.0	14.5	14.5	17.5
	Pentagastrin	17.5	18.5	18.5	19.5	16.5	20.0	16.0	19.5
Mean	Saline	17.2	17.2	17.2	18.9	20.0	20.0	20.6	20.2
S.D.		7.59	7.28	8.34	7.46	7.55	7.97	8.45	7.28
Mean	Pentagastrin	21.6	23.5	23.7	24.5	24.7	25.8	25.3	25.6
S.D.		8.33	7.31	8.17	8.59	8.72	7.91	9.11	8.91

Appendix 11.4 Effect of atropine (15 $\mu g.kg^{-1}$ i.v.) on lower oesophageal sphincter pressure

Sheep No.	Basal LOS pressure (mmHg)	LOS Pressures After Atropine Injection (mmHg)						
		1 min	5 mins	10 mins	15 mins	20 mins	25 mins	30 mins
Z981	11.5	11.5	11.5	11.0	10.0	8.0	11.0	11.5
645B	15.0	15.5	15.5	15.0	8.5	12.5	13.0	15.0
567E	12.5	12.5	13.0	12.5	10.0	11.5	12.5	11.5
566E	13.5	13.5	12.5	12.5	14.0	12.0	12.5	12.0
518E	10.0	10.5	9.0	11.0	15.0	11.5	14.5	15.0
Z981	9.0	9.0	9.5	9.0	9.0	9.0	9.0	9.0
Z650	8.5	8.5	9.0	9.5	9.5	9.0	8.5	8.5
Mean	11.42	11.57	11.42	11.5	10.85	10.5	11.57	11.78
S.D.	2.4	2.49	2.43	2.04	2.56	1.77	2.18	2.56

Appendix 11.5 Effect of atropine (30 $\mu g.kg^{-1}$ i.v.) on lower oesophageal sphincter pressure

Sheep No.	Basal LOS pressure (mmHg)	LOS Pressures After Atropine Injection (mmHg)						
		1 min	5 mins	10 mins	15 mins	20 mins	25 mins	30 mins
567E	12.5	11.0	11.5	12.0	13.0	12.5	12.5	12.5
645B	10.0	8.5	8.5	9.0	9.5	9.5	10.0	10.5
566E	15.0	15.0	13.5	14.0	14.5	14.5	15.0	15.5
Z650	13.5	13.0	13.0	13.5	12.5	12.5	13.0	14.0
594E	13.5	10.0	12.5	15.0	11.0	11.5	12.5	14.0
Mean	12.9	11.5	11.8	12.7	12.1	12.1	12.6	13.3
S.D.	1.85	2.54	1.98	2.33	1.91	1.81	1.78	1.89

Appendix 11.6 Effect of propranolol hydrochloride (0.05 $mg.kg^{-1}$ i.v.) on lower oesophageal sphincter pressure (after atropine, 30.0 $\mu g.kg^{-1}$)

Sheep No.	Injection	Basal LOS pressure (mmHg)	LOS Pressures After Propranolol Hydrochloride Injection (mmHg)						
			1 min	5 mins	10 mins	15 mins	20 mins	25 mins	30 mins
498E	Saline	20.0	20.0	23.0	22.5	24.0	23.0	20.0	25.0
	Propranolol	24.0	23.0	19.0	19.0	22.0	23.0	25.0	25.0
528E	Saline	16.0	16.5	16.0	15.0	16.0	15.0	16.0	18.0
	Propranolol	16.0	16.0	16.5	17.0	15.0	15.5	14.5	14.0
550E	Saline	26.0	26.0	25.5	26.0	25.5	25.5	22.5	21.0
	Propranolol	20.0	25.5	20.0	20.0	19.5	18.0	18.0	17.5
514E	Saline	15.0	14.5	14.5	15.0	14.0	15.0	16.0	14.0
	Propranolol	14.0	14.0	15.0	15.0	13.0	12.0	12.0	11.5
510E	Saline	17.5	17.0	21.0	18.0	17.5	16.5	15.0	15.0
	Propranolol	15.0	13.0	12.5	12.5	13.0	10.5	12.0	13.0
Mean	Saline	18.9	18.8	20.0	19.3	19.4	19.0	17.9	16.6
S.D.		4.4	4.5	4.7	4.8	5.06	4.9	3.2	4.5
Mean	Propranolol	17.8	18.3	16.6	16.7	16.5	15.8	16.3	16.2
S.D.		4.1	5.6	3.02	3.03	4.06	5.0	5.4	5.4